CONFERENCE ON DISARMAMENT

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Working Paper on Challenge Inspections/Inspections on Request

I. Introduction

1. The verification system flowing from an arms limitation treaty encompasses a group of rules, methods, procedures and verification techniques aimed at guaranteeing that any Contracting Party comply with its obligations. In other terms: verification is a human activity for tangible purposes under given parameters, so that hidden intentions can never be detected.

2. As a human invention, verification is far from being 100 per cent accurate: absolute verification or a perfect verification system remain a utopian exercise. On the other hand, verification is not enough in itself to deter any Contracting Party from violating its obligations. A set of credible sanctions is needed to match the detection risk. Generally speaking, it seems easier to verify the complete elimination of something than its partial banning. However, the difficulties arise "au fur et à mesure" the ambiguities surrounding the main obligations or the complexities of the subject matter increase. In any event, the verification of the arms limitation treaties is a kind of confidence-building measure intended to allay the fears among the States involved and provide them with a reasonable degree of reciprocal security.

II. Objectives and Structure

3. The multilateral Covention on the complete and effective prohibition of chemical weapons and on their destruction aims at two main objectives: (i) chemical disarmament which implies, grosso modo, the elimination of all existing arsenals of chemical weapons as well as all chemical weapon production facilities of the States Parties; and (ii) peaceful uses of chemical industry, including the prevention of the development of chemical weapon capabilities.

4. To achieve these objectives the international verification system, as it appears in the "Rolling Text" (document CD/1046), currently under elaboration, has already a structure based on two main pillars: (i) the verification of compliance with the provisions pertaining to chemical disarmament (articles III, IV and V, in accordance with annexed texts); and (ii) the

verification of compliance with the provisions dealing with activities not prohibited under the Convention (article VI, paras. 1 and 2, in accordance also with annexed texts). In addition, both are strengthened by a complementary mechanism specially aimed at clarifying any doubt or concern about compliance with those obligations, the short-notice challenge inspection/inspection on request being its most outstanding component (article IX).

III. Challenge Inspections/Inspections on Request

5. This kind of inspection is one of the main features of the international verification system. Besides the fact already mentioned that absolute verification is beyond human possibilities, routine verification in all facilities relevant to the Convention implies an "unmanageable burden" on the Technical Secretariat with direct repercussion in its size and financing of the numerous verification activities.

6. Thus, the challenge inspection/inspection on request constitutes an indispensable device for complementing a set of reasonable and affordable verification measures related to articlves IV, V and VI. In this connection, challenge inspection/inspection on request, could be taken as a confidence-building measure or a "safety net" in order to safeguard confidence and prevent threats against the security of the States Parties to the Convention.

7. Bearing in mind the principle of non-discrimination as recognized by the 1988 session of the United Nations Disarmament Conference and endorsed by the United Nations General Assembly, the implementation of this complementary mechanism should not give place to any kind of discrimination between the requesting and the requested States Parties. On the other hand, every State Party should accept as a basic principle that all Contracting Parties intend to behave correctly and that they will comply faithfully with the provisions of the Convention. In this line of thought, the right of the requesting State must not override the right of the requested State. "Bonna fide" of the requesting and requested States Parties is of utmost importance so as to preclude abuse of this kind of inspection. Accordingly, the notion of suspicion should be excluded at the beginning.

8. Furthermore, the right to set-off a challenge inspection/inspection on request, is not unlimited or at least it should be restricted at the beginning precisely because its purpose is to demonstrate compliance with the obligations and not to prejudge suspected activities in the requested State. Additionally, the protection of confidential information also constitutes another barrier to free access any time to the declared facilities or areas for verification purposes. So, the notion that this kind of inspection is to be "conducted anywhere without delay" seems premature if it is mandatory from the very beginning and the obligatory presence of observers of the requesting State risks to question not only the reliability of the international verification system, but also the principle of non-discrimination among States involved.

9. One way to solve this problem, without placing in jeopardy the universal acceptance of obligatory challenge inspection/inspection on request, could be to conduct this kind of inspection in two stages. In the first one, technically oriented and strictly confidential, all the steps will remain

within the competence and powers of the Director-General. Upon receipt of a request coming from a State Party, he should inform, within the following 24 hours, the requested State as well as the Executive Council. Then, in the following 24 hours he should designate an inspection team which should immediately reach the territory of the requested State and have access to any facility or installation relevant to the request, but not yet "anywhere".

10. However, if the inspection team does not succeed in clarifying the existing doubts or concerns as they appear in the request, then the Executive Council should meet within the following 48 hours to consider the extension of the mandate to "anywhere" within the scope of the request in order to make a final assessment about the situation and whether or not the Convention is being violated. This second stage would be of a politial nature and utilized as a weapon of last resort in the field of inspections, before further action - including possible sanctions - is taken.

11. There is no doubt that a request has to be as precise as possible. Although access must never be refused as a matter of principle, there is no question in the first stage of conducting a challenge inspection/inspection on request beyond the parameters of the request itself (nature of the suspected non-compliance, the specific site to be inspected and matters on which reassurance is required). That is not the case for the second stage where further access without any restriction relevant to the request should be the main component, given its highly political implications.

12. The principles of non-discrimination and good faith among requesting and requested States Parties might also render it advisable to include in the Convention provisions to discourage any abuse of the right of States Parties to request challenge inspection/inspection on request. Sanctions could be envisaged in case a State Party reiterates requests which are groundless.

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LETTER DATED 16 MAY 1991 FROM THE DEPUTY PERMANENT REPRESENTATIVE OF AUSTRIA TO THE SECRETARY-GENERAL OF THE CONFERENCE ON DISARMAMENT TRANSMITTING A STUDY ENTITLED "DETECTION OF INHIBITORS OF THE ENZYME ACETYLOCHOLINE ESTERASE OVER LONG DISTANCES USING OPTICAL FIBERS"

I have the honour to forward to you a copy of an Austrian study entitled "Detection of inhibitors of the enzyme acetylocholine esterase over long distances using optical fibers" and I would ask you to be kind enough to circulate it as an official document of the Ad Hoc Committee on Chemical Weapons as well as of the Conference on Disarmament.

(Signed) Christian Strohal
Deputy Permanent Representative
to the Conference on Disarmament

GE.91-60871

Project

Detection of inhibitors of the enzyme acetylcholine esterase over long distances using optical fibers

AUSTRIAN SCIENTIFIC COUNCIL FOR THE IMPLEMENTATION OF THE CHEMICAL WEAPONS CONVENTION

final report, April 1991

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Abstract

Many pesticides (such as organophosphates or carbamates) and most nerve agents used in chemical warfare (such as Tabun, Sarin or VX) are potent inhibitors of a group of enzymes present in humans and animals, called choline esterases. The activity of one of these esterases, the acetylcholine esterase (AChE), is essential for the function of the nerve system. An inhibition of the AChE in humans leads to sweating, headache and running nose in the case of a minor contamination. If the AChE is completely inhibited, cramps, paralyzation of the breath center and finally the death are the result.

Pesticides are commonly found in ground water and drinking water due to their extensive use in agriculture as crop protection agents. The production and the transport of these substances represents another area of possible contamination of humans and of the environment (i.e. Cordoba, Mexico, May 1991). As recent political developments have shown, the use of chemical weapons still threatens millions of people. The rapid and accurate detection of inhibitors of the acetylcholine esterase therefore represents an important preventive procedure for the protection of humans, animals and environment.

We have developed a new method for on-line detection of inhibitors of the AChE in a continuous flow system. The presence of inhibitors is detected optically through fibers. Therefore, the sensing part of the apparatus can be as far as 1000 meters and more away from the detecting part. The detection principle combines low detection limits (for example 10 nM of the potent reversible inhibitor BW 284 C51) with remote sensing, cheap and robust solid state technology (light emitting diodes as light sources and photodiodes as light detectors) and ease of operation. The detection limits for the presence of inhibitors of the AChE can be enhanced by one to two orders of magnitude. This can be achieved through an optimization of the electronical, chemical and optical components of the system without major increase of the costs of the system.

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THE DETECTION OF INHIBITORS OF THE ENZYME ACETYLCHOLINE ESTERASE OVER LONG DISTANCES USING OPTICAL FIBERS

1. The detection of inhibitors of the enzyme acetylcholine esterase: concern and pertinence [1-10]

Acetylcholine is the most important chemical transmitter in the nerve system of humans and animals (Fig.1). If a sense-organ is stimulated by an external event, the excitation is transformed into bioelectrical impulses which are transmitted through the nerve fibers to the central nerve system and/or the brain.The nerve fibers consist of many nerve cells which are connected to each other by a slit, called synapse. The electrical information is transmitted chemically across the synapse between two nerve cells. The transmitter substance in this chemical reaction is the acetylcholine. It is released at one side of the synapse through the excitation arising from the electrical impulse of one nerve cell. Acetylcholine then diffuses across the slit to the other side of the synapse and excites the next nerve cell. In fact, the transmission of the stimulation information of the sensing organ can be viewed as succeeding reaction of many electrical and chemical transmission steps.

Fig. 1

After excitation of the nerve cell, the acetylcholine molecule has to be destroyed in order not to excite continuously the nerve cell. This deactivation is performed by the enzyme acetylcholine esterase which hydrolyzes the acetylcholine into choline and acetic acid. An inhibition of the enzyme acetylcholine esterase results in an incomplete deactivation of the transmitter acetylcholine. The consequences in humans are sweating, headache and running nose in the case of a minor inhibition. If the acetylcholine esterase is completely inhibited, cramps, paralyzation of the breath center and finally the death are the result. The inhibition of the enzyme may be reversible or irreversible, depending on the desired area of application. Since many inhibitors of the acetylcholine esterase are commonly used in civil as well as military tasks, the importance of a detection method for these substances becomes obvious.

Several classes of substances are potent inhibitors of the enzyme acetylcholine esterase: Organophosphates, carbamates, quarternary ammonium ions and methanesulfonates. Organophosphorus inhibitors of the acetylcholine esterase have found application in two main areas: as pestizides in agriculture (i.e. E 605) and as chemical weapons (i.e. Sarin, Tabun). Chemical warfare agents and pesticides cannot be separated strictly since some organo-phosphorus compounds have served many years both purposes. The main difference between the pesticides and the chemical weapons is the choice of the substitutents at the phosphorus atom during syntheses. For example, fluoride has been used as a substituent in chemical weapons to enhance the speed of inhibition of acetylcholine esterase. Carbamates are used extensively in the crop protection. Quarternary ammonium ions and carbamates both have served in medicine as parasympathomimetic agents.

2. The determination of inhibitors of the enzyme acetylcholine esterase

The most widely-used methods for the determination of organophosphorus and carbamate pesticides are high-pressure liquid chromatographie (HPLC), gas chromatographie (GC) and mass spectroscopy (MS) [11]. All of these methods require a tedious sample pretreatment, sophisticated instrumentation and are expensive.

To detect inhibitors of the enzyme acetylcholine esterase (AChE) it is advantageous to use the enzyme itself. For this purpose the activity of the enzyme is measured in the presence of an inhibitor after an incubation time of typically 30 minutes. Many methods have been described which differ in the way in which the activity of the enzyme is determined [11,12].

The natural substrate of the enzyme is acetylcholine which is cleaved by acetylcholine esterase to give acetate and choline (Equ. 1). In a radiometric assay acetylcholine was labelled with tritium (H^3) or with carbon C^{14} at the acetyl group. After reaction with the enzyme, the acetic acid produced was separated and its radioactivity was measured [11,12]. The problems of this method are the radioactive labelling of the acetylcholine, the radioactive waste, which has to be dispose and the radioactivity itself.

$$\text{acetylcholine} + H_2O \xrightarrow{\text{AChE}} \text{acetate} + \text{choline} \qquad \text{(Equ. 1)}$$

In electrochemical methods the production of acetic acid (Equ. 1) was followed with a pH electrode [12] or with a pH sensitive field effect transistor (ISFET) [13]. Very often, the reaction of acetylcholine esterase is coupled to a second enzymatic reaction. The enzyme choline oxidase (ChOD, EC 1.1.3.17) oxidizes choline and as products betaine and hydrogen peroxide are formed (Equ. 2). The hydrogen peroxide can be detected amperometrically with a platinum electrode [14,15].

$$\text{choline} + 2\,O_2 + H_2O \xrightarrow{\text{ChOD}} \text{betaine} + H_2O_2 \qquad \text{(Equ. 2)}$$

In an other method, acetylthiocholine iodide has been used instead of acetylcholine and the formation of thiocholine was monitored potentiometrically [12].

Synthetic enzyme substrates are very often used in optical methods for the determination of inhibitors of acetylcholine esterase. The substrates and products usually show different colours and the colour changes occuring during the enzymatic reaction can be detected photometrically. A typical example is the yellow substrate indophenyl acetate, which is hydrolyzed by cholinesterases producing an intensely blue product [11].

Fluorescent dyes have also been used in detecting inhibitors of acetylcholine esterase. Indoxylacetate or resorufinacetate show a strong fluorescence after hydrolysis by the enzyme whereas the substrates do not show fluorescence. Unfortunately the excitation wavelengths of most of these compounds are in the ultraviolet region of the light [12].

Other colorimetric methods are based on complex reactions to produce a dye whose absorption is measured. The method from Ellman is well known [12]: Acetylthiocholine is hydrolyzed by acetylcholine esterase and the thiocholine formed reacts with 5,5´-dithiobis-(2-nitrobenzoate) to give a yellow dye.

Immunological reactions have also been used for the determination of pesticides. In an immunoassay for paraoxon, the high selectivity and sensitivity of an antigen/antibody reaction was coupled with the specificity of an enzymatic reaction [11]. Although this method was highly sensitive for paraoxon, it could be used for the detection of this single pesticide only. In addition it was very expensive and time consuming.

In this report we present a novel optical method for the determination of inhibitors of acetyl- and butyrylcholine esterases. We use synthetic enzyme substrates which are yellow in aqueous solution and are converted into blue products by choline esterases. In

absence of an inhibitor, all substrate is hydrolyzed by the enzyme to give the blue product. In presence of an enzyme inhibitor the amount of blue dye formed is reduced. This causes a change in colour, which may serve as an alarm for the presence of enzyme inhibitors for example in a drinking water reservoir. As the enzyme substrate is the indicator itself, no additional chemical reactions (and therefore no additional reagents) are required. The enzymes are immobilized covalently on glass or polymer particles and can be used over several days continuously in an enzyme reactor.

The blue colour of the products allows the detection with the help of a yellow light emitting diode (LED) as the light source. Cheep LED´s with high light out-put are available for longer wavelengths of the visible spectrum only. The use of optical fibers makes tit possible to perform measurements ove- large distances and at inaccessible places. Additionally, in this work we use a complete new type of a photometer, which is equipped with inexpensive components like LED´s as light sources, photodiodes as detectors and plastic fibers. With this instrument, a simple and cheap instrumentation is possible, in contrast to the expensive commercial photometers.

If lasers are used as light sources in connection with optical glass fibers, measurements can be performed over distances of at least several hundred meters. The procedure was designed as an on-line method for continuous measurement. As all substances which inhibit the enzymes acetylcholine esterase and butyrylcholine esterase can be detected, a kind of a sum of parameters is measured. We assumed that it is of interest to detect any substance which is harmful to the human nerve system and therefore to the whole organism. We were not looking for a method allowing the identification or quantification of pesticides or nerve gases in drinking or ground water. Of course the method shows different sensitivities towards the various inhibitors due to the different inhibiting properties of the substances.

3. Experimental

3.1. Materials

The enzymes acetylcholine esterase (EC 3.1.1.7; type VI-s, from electric eel) with a specific activity of 200 U/mg and butyrylcholine esterase (EC 3.1.1.8; from horse serum) with a specific activities of 360 or 308 U/mg were obtained from Sigma Chemie GmbH (Deisenhofen, FRG).

The synthetic enzyme substrates were Substrate A (with an acetoxy group) and Substrate B (with a butyroxy group). These compounds are proprietary substances.

The enzyme inhibitors paraoxon (diethyl-p-nitrophenylphosphate, E 600; 95 % liquid), pyridostigmine bromide and 1,5-bis(allyldimethyl-ammoniumphenyl)-pentane-3-one dibromide (BW 284 C51) as well as the reactivating reagent pyridine-2-aldoxime methoiodide (2-PAM) were purchased from Sigma Chemie GmbH (Deisenhofen, FRG).

Controlled pore glass Aminopropyl-CPG-170-A (120 - 200 mesh; with 0.2 mmol amino groups/g) was from Fluka Chemie AG (Buchs, Switzerland). Polymer beads with oxiran groups on the surface (300 µmol/g; particle size 50 - 200 µm) were obtained from Riedel-de Haen AG (Seelze, FRG). Nylon nets (with pore sizes of 30 and 60 µm) for sealing the enzyme reactors were purchased from Reichelt Chemietechnik GmbH + Co (Heidelberg, FRG).

All other chemicals used were of analytical-reagent grade.

3.2. Immobilization of the enzymes

The enzymes acetylcholine esterase from electric eel and butyrylcholine esterase from horse serum were used because they are available with high specific activities - a fact that is essential for the immobilizazion of the enzymes. We tried to immobilize both enzymes on different materials, such as controlled pore glass (modified with aminopropyl groups) or polymer particles (with oxiran groups on the surface). The glass or polymer beads

with the immobilized enzyme were filled in enzyme reactors with a reactor length of 1 cm and a volume of approximately 70 µl. The enzyme reactors were sealed on both ends with nylon nets and stored in buffer solution at 4 °C in a refrigerator untill they were used.

3.2.1. Immobilization on aminopropyl-CPG with carbodiimide

3.1 mg of acetylcholine esterase (620 units) or 2.2 mg of butyrylcholine esterase (677 units) was dissolved in 3.0 ml of 0.1 M phosphate buffer pH 7.0. After the addition of 0.2 g of aminopropyl-CPG and 50 mg of N-ethyl-N´(3-dimethylaminopropyl)-carbodiimide hydrochloride (EDC), the solution was stirred for one hour at room temperature and stored at 4 °C for 24 hours in a refrigerator. The CPG was washed with cold 0.1 M phosphate buffer pH 7.0 and stored in the same buffer at 4 °C.

3.2.2. Immobilization on aminopropyl-CPG with glutaraldehyde

This immobilization method was adapted from a published procedure [7]. One ml of a 2.5 % solution of glutaraldehyde in 0.1 M phosphate buffer pH 7.0 was added to 0.2 g of aminopropyl-CPG. Before and after addition of the CPG, nitrogen was bubbled through the glutaraldehyde solution. After standing for one hour, the CPG was washed with water. 3.47 mg of acetylcholine esterase (694 units) or 1.8 mg of butyrylcholine esterase (648 units) were dissolved in 3.0 ml of cold 0.1 M phosphate buffer pH 6.0. The preactivated CPG was added to the enzyme solution and left to stand in a refrigerator at 4 °C for 2.5 or for 24 hours, respectively. The CPG was washed with cold 0.1 M phosphate buffer pH 6.0 and stored in the same buffer at 4 °C.

3.2.3. Immobilization on polymer particles via oxiran groups

3.40 mg acetylcholine esterase (646 units) or 1.03 mg of butyrylcholine esterase (317 units) were dissolved in 3.0 ml of 1 M potassium phosphate buffer pH 7.5. Then 2.53 or 1.59 mg of VA-Epoxy Biosynth (polymer beads with oxiran groups on the surface) were added, respectively. After two days of storage in a refrigerator at 4 °C, the polymer beads were washed with 0.1 M phosphate buffer of pH 7.4 and stored in 0.1 M phosphate buffer pH 7.0 at 4 °C.

3.3. The enzyme substrates

The enzyme substrates (Substrate A and Substrate B) are both highly soluble in water and water/solvent mixtures. The substrate solutions show a yellow colour and the absorption maxima in destilled water are at about 390 nm, whereas the products of the enzymatic hydrolysis are blue with absorption maxima at about 580 nm (Fig. 2).

The substrates display a good stability in destilled water, whereas they are less stable in solvents like ethanol or dioxan. In addition, they are also not stable in aqueous buffered solutions. Only in mixtures of water and cellosolve (2-ethoxy-ethanol) the stability of the substrate was highly improved. Unfortunately cellosolve led to a rapid decrease in enzyme activity which may be due to inhibition or denaturation of the enzyme. Therefore only substrates dissolved in distilled water were used. Figs. 3 and 4 show the spontaneous hydrolysis of the substrates in distilled water as a function of time and temperature. The solid substrates, which are orange-red cristalls, were stored below 0 °C under exclusion of moisture. No changes were observed over a period of several weeks.

Figs. 2, 3 & 4

3.4. The inhibitors

Numerous inhibitors of the enzymes acetyl- and butyrylcholine esterase are known [8]. We chose three representative examples of the most important substance groups: an organophosporus compound (paraoxon), a carbamate (pyridostigmine bromide) and a quarternized ammonium ion (BW 284 C51).

From paraoxon (liquid, 95 %) a 0.16 M stock solution was prepared in acetone. Aqueous 160 µM stock solutions were obtained by diluting this solution with destilled water. Pyridostigmine bromide and BW 284 C51 were cristalline and were dissolved in distilled water directly. The aqueous carbamate and paraoxon solutions were prepared fresh every day. For decontamination, the pesticide solutions and the contaminated materials were treated with 0.1 M solution of potassium hydroxide to hydrolyze paraoxon and pyridostigmine. BW 284 C51 was destroyed with the help of an alkaline solution of potassium permanganate.

3.4.1. Paraoxon (Diethyl-p-nitrophenylphosphate; E 600)

Paraoxon (Fig. 5a) is a well known example of the organophosphorus pesticides. The oral lethal dose LD50 is 1.8 mg/kg for rats and 5 mg/kg for men. Paraoxon is also a metabolite of the less toxic parathion (E 605) which has found wide application as pesticide. It is transformed into the highly toxic paraoxon in the body and the lethal dose is 300 - 400 mg for an adult man [2,4,17].

Paraoxon inhibits both, acetyl- and butyrylcholine esterase irreversibly by phosphorylation of the active site of the enzyme. Although the spontaneous regeneration of the enzyme is very slow (with half-life times of the phosphorylated enzyme in the order of days), it is possible to reactivate the enzyme with substances like pyridine-2-aldoxime-methoiodide (2-PAM, Pralidoxim). This reactivation is possible before the diethylphosphate group is partially hydrolyzed ("aging" of the phosphorylated enzyme) [1,3]. Since aging is very slow in the case of paraoxon as the inhibitor, the reactivation with 2-PAM occurs fast [11,13,14].

Fig. 5

3.4.2. Pyridostigmine bromide (3-Dimethylaminocarbonyloxy-N-methyl-pyridinium bromide; Mestinon)

Pyridostigmine bromide (Fig. 5b) was chosen, because it also inhibits both, acetyl- and butyrylcholine esterases [18]. The inhibition occurs through carbamylation of the enzyme, but the hydrolysis of the ester formed is fast when compared with the phosphorylated enzyme. The half-life times of carbamylated enzymes are in the order of 1 to several hours (depending on the type of enzyme) [1]. Pyridostigmine has been used as a so called parasympathomimetikum in medicine [2,3].

3.4.3. BW 284 C51 (1,5-bis(4-Allyldimethyl-ammoniumphenyl)-pentane-3-one dibromide)

The compound BW 284 C51 (Fig. 5) is a typical representative of inhibitors of the type of substituted ammonium ions. It is a potent reversible inhibitor of acetylcholine esterase and has a LD50 of 2.1 mg/kg for mice. It can bind to the anionic binding site of the enzyme and it has been used as a selective inhibitor for acetylcholine esterase in medical research [18].

3.5. Buffer solutions

Buffering of the substrate solutions in the enzyme reactor is essential because the enzyme activity and the enzyme inhibition are both pH dependent [6,17]. The addition of sodium

chloride and magnesium chloride may enhance the lifetime of the immobilized enzyme considerably. It was reported, that in the presence of 12 µM $MgCl_2$ and 45 mM NaCl the lifetime was three times higher than in absence of these salts [17]. Therefore all experiments were performed with buffers containing similar salt additions. Depending on the mixing ratio of buffer:substrate:inhibitor solutions we used 0.2 M sodiumphosphate buffer containing 180 mM NaCl and 48 µM $MgSO_4$ for a ratio of 1:1:2 and 0.1 M sodiumphosphate buffers containing 90 mM NaCl and 24 µM $MgSO_4$ for a ratio of 1:1:1. Buffer and substrate solutions had to be separated because the substrate is not stable in buffered solutions especially at pH > 7.

4. Measurements with an UV/VIS-spectrophotometer

The experimental set-up is shown in Fig. 6. The enzyme substrate solutions are mixed with buffer and inhibitor solution via a peristaltic pump and are pumped through an enzyme reactor. Here, the yellow substrate is hydrolyzed by the enzyme and the blue product is formed. In presence of an inhibitor the formation of this blue product is reduced because the enzyme is inhibited. This causes a change in absorbance at 580 nm which can be monitored with an UV/VIS-spectrophotometer.

Fig. 6

The spectrophotometer was a Perkin-Elmer Lamda 5 UV/VIS Spectrophotometer (Perkin-Elmer & Co GmbH, Überlingen, FRG). A flow-through cell with a light path of 1.5 mm from Hellma GmbH & Co (Müllheim, Baden, FRG) was used for the continuous flow measurements. For investigating the substrate stabilities 1x1-cm quartz cuvettes were used. The spectrophotometer allowed to measure absorption spectra in the flow-through cell which was advantageous in the beginning of the studies.

The peristaltic pump was a Gilson Minipuls 3 (Gilson Medical Electronics, Villiers-le-Bel, France) or a MV-CA4 from ISMATEC GmbH (Wertheim, Mondfeld, FRG).

The inhibitor solution represents a drinking or ground water reservoir, from which a sample is drawn continuously. In absence of an inhibitor, destilled water was used instead of the inhibitor solution. The mixing ratio was 1:1:1 for substrate:buffer:inhibitor at the beginning and was changed to 1:1:2 to increase the sensitivity of the method later on.

All measurements were performed at room temperature, i.e. 25 ± 1 °C.

4.1. Measurements with acetylcholine esterase immobilized on aminopropyl-CPG

Immobilization of acetylcholine esterase on aminopropyl-CPG with carbodiimide or glutaraldehyde yielded a material with high enzyme activity and good stability. After storage in a refrigerator (5 °C) for four months, no considerable loss in enzyme activity was observed. An enzyme reactor with acetylcholine esterase immobilized on aminopropyl-CPG with carbodiimide could be used for more than 50 hours (over a period of two weeks) in continous flow and was exposed to numerous inhibition tests with paraoxon, pyridostigmine or BW 284 C51. Enzyme reactors which were not in use, were stored at 5 °C in a refrigerator.

Fig. 7 shows the pH-dependence of the hydrolysis of Substrate A with a fresh prepared enzyme reactor. A broad pH-optimum from approximately pH 5.5 to 9.0 can be observed, which is due to a high enzyme activity in the enzyme column. A change in the flow-rate did not affect the results in the range of 0.5 to 1 ml/min.

Fig. 7

4.1.1. Inhibition with paraoxon

The inhibition of acetylcholine esterase with paraoxon is shown in Fig. 8. The changes in absorbance were measured after a time of 21 minutes. Paraoxon is an irreversible inhibitor of the enzyme. Therefore the concentration of the blue product decreases continuously with time since more enzyme gets inhibited. After the inhibition reaction, the reactivation was performed with 0.1 mM solution of 2-PAM and the enzyme activity was restored (Fig. 9). The detection limit was about 0.2 µM of paraoxon using this method.

Figs. 8 & 9

Investigation of the pH-dependence of the inhibition reaction showed an optimal pH-range of approximately 7.5 to 8.5 (Fig. 10). The effect of the flow-rate on the inhibition is shown in Fig. 11. The signal changes increased in the range of 0.4 to 2.0 ml/min.

Figs. 10 & 11

4.1.2. Inhibition with pyridostigmine bromide

The inhibition of acetylcholine esterase immobilized on aminopropyl-CPG with pyridostigmine bromide is shown in Fig. 12. The changes in absorbance were measured after 21 minutes, although - in contrast to paraoxon inhibition - a steady-state response is established after some time. The limit of detection was about 1 µM of pyridostigmin. The regeneration times were 15 to 30 minutes and depended on the concentration of the inhibitor. Reactivation of the enzyme occured spontaneously and was not influenced by substances like 2-PAM.

Fig. 12

4.1.3. Inhibition with BW 284 C51

Fig. 13 depicts the inhibition of acetylcholine esterase immobilized on aminopropyl-CPG with the reversible inhibitor BW 284 C51. A steady-state was observed after a time of about 5 to 15 minutes (Fig. 14). The regeneration times varied from 8 to 25 minutes (depending on the concentration of the inhibitor). The detection limit was approximately 10 nM of BW 284 C51.

Figs. 13 & 14

An increase in the flow-rate caused an increasing sensitivity (Fig. 15). The reason may be the high affinity of the enzyme to the inhibitor. Because of the two positive charges BW 284 C51 is more readily bound to the anionic binding site of the enzyme than the enzyme substrate.

Fig. 15

4.2. Measurements with acetylcholine esterase immobilized on VA-Epoxy-Biosynth

VA-Epoxy-Biosynth polymer beads with immobilized acetylcholine esterase also showed high enzyme activity and were suitable for inhibition tests. The inhibition of acetylcholine esterase with paraoxon is shown in Fig. 16. The signal changes were again measured after 21 minutes. The limit of detection was approximately 0.5 µM paraoxon and the reactivating times ranged from 10 to 20 minutes.

Fig. 16

4.3. Measurements with immobilized butyrylcholine esterase

The immobilization of butyrylcholine esterase was less successful than the immobilization of acetylcholine esterase. Enzyme reactors which were filled with butyrylcholine esterase immobilized on aminopropyl-CPG with carbodiimide showed almost no enzyme activity. Substrate B and Substrate A were not hydrolyzed. Immobilization of butyrylcholine esterase on aminopropyl-CPG with glutaraldehyde yielded better results. Fig. 17 shows the pH-dependence of the hydrolysis of Substrate B. A distinct pH-optimum of pH 7 can be observed. The signal was dependent on the flow-rate (Fig. 18) which indicates a low enzyme activity on the carrier material. At pH 7.5 and at a flow-rate of 1.8 ml/min only 50 % of the substrate were converted into the blue product. Enzyme reactors with butyrylcholine esterase immobilized on VA-Epoxy-Biosynth also showed low enzyme activities. At pH 7.5 and at a flow-rate of 1.8 ml/min only 25 % of the substrate were hydrolyzed.

Immobilized butyrylcholine esterase was inhibited by paraoxon or pyridostigmine, but almost no reactivation of the inhibited enzyme was observed. After inhibition with paraoxon it was, for example, not possible to reactivate the enzyme with 2-PAM. Only a small part of the enzyme activity was restored. Even inhibition with pyridostigmine was irreversible. Therefore it was not possible to determine detection limits or calibration graphs because each enzym reactor could be used for one inhibition test only before it had to be replaced. For that reason enzyme reactors with immobilized butyrylcholine esterase were not suitable for investigation and test purposes. Nevertheless, they could be used in a pesticide alarm system, where a single response is sufficient.

Figs. 17 &18

5. Measurements performed with a fiber optic photometer and a laser

Fig. 19 shows the experimental set-up for the measurements with the fiber optic photometer. The arrangement was the same as described in Chapter 4. Instead of the UV/VIS spectrophotometer, the 3-Lambda-Photometer from the Institute for Optical Sensors (Joanneum Research, Graz, Austria) was used. This photometer is equipped with solid-state components, such as light-emitting diodes (LED´s) as light sources and photodiodes as detectors. The photometer was connected to the flow-through cell with plastic fiber light guides. The flow-through cell was made of stainless steel plates with a teflon spacer in the centre. The light path through the cell was 2.5 mm long. In addition, the photometer signal was recorded with a Gould BS-273 three-channel recorder (Gould Bryans Instruments Ltd., Mitcham, Surrey). For all measurements, a yellow LED was used as the light source.

For measurements with the laser as the light source, a Helium-Neon-Laser (PMS Electro-Optics, Boulder, Colorado, USA) with an emission wavelength of 612 nm was used. The detector was a lightwave multimeter 8153A equipped with an optical head 81520A from Hewlett-Packard (Vienna, Austria). Plastic fibers with a thickness of 1000 µm and glass fibers with a thickness of 125 µm, as well as optical fiber couplers, were obtained from Hirschmann (Vienna, Austria).

Fig. 19

5.1. Measurements with the 3-Lambda-Photometer

For testing the 3-Lambda-Photometer the sample flow passed first the flow-through cell of the UV/VIS spectrophotometer and second the flow-through cell of the fiber-optic photometer in order to perform simultaneous measurements with two detectors. For most of the experiments, an overall length of 10 meters of plastic fibers was used. It was shown, that almost no differences could be observed between the results obtained with both photometers, although the fiber-optic photometer did not use a reference beam.

The inhibition of acetylcholine esterase (immobilized on polymer beads) with paraoxon is shown in Fig. 20. The results are identical with that from Fig. 16, which were obtained with the spectrophotometer. Only minor differences can be observed between the two calibration graphs.

Fig. 20

The inhibition of acetylcholine esterase immobilized on aminopropyl-CPG with pyridostigmine is shown in Fig. 21. The calibration graph is almost identical with that from Fig. 12, which was measured simultaneously.

Fig. 21

A total of 90 meters of plastic fibers was used in the measurement shown in Fig. 22. The main difference between the measurements with 10 and 90 meters of plastic fibers was the smaller signal/noise ratio when 10 meters of plastic fibers were used. The limit of detection was approximately 30 nM for BW 284 C51. However, a length of 100 meters of plastic fibers appeared to be the upper limit when using the 3-Lambda-Photometer with a LED as the light source.

Fig. 22

5.2. Measurements with a laser

The inhibition of acetylcholine esterase immobilized on aminopropyl-CPG with BW 284 C51 (Fig. 22) was repeated with a He-Ne-laser as the light source. The result (Fig. 23) is comparable to that obtained with the 3-Lambda-Photometer despite some differences in the central part of the calibration graph. 90 meters of plastic fibers were used again, but the signal/noise ratio was not as high as with the 3-Lambda-Photometer. This may be due to problems with coupling the light of the laser into the fiber and to laser instabilities. Little changes in the connection of fiber coupler and laser resulted in high signal fluctuations. The detection limit was approximately 50 nM for BW 284 C51.

Fig. 23

Nevertheless, these measurements demonstrate the usefulness of a laser as a light source. With the high output of a laser combined with the use of glass fibers it is possible to measure the signal changes which occure during the inhibition of choline esterases over large distances. A detection of inhibitors over a distance of about 100 meters is possible with the orange He-Ne-laser and with 1000-µm plastic fibers. With the same laser and 125-µm glass fibers measurements can easily be performed over a distance of about 1 kilometer.

6. Discussion

The results demonstrate, that it is possible to detect inhibitors of the enzymes acetyl- and butyrylcholine esterase under the applied conditions on-line, i.e. continuously over a period of several days. The use of optical fibers allows measurements over large distances. With the novel and robust fiber-optic 3-Lambda-Photometer, which is equipped with light emitting diodes, photodiodes and optical plastic fibers, it is possible to measure over a distance of approximately 50 meters from the place where the samples are drawn. Distances of up to one kilometer can be acchieved if lasers and glass fibers are used.

The detection limits for paraoxon of 200 nM (= 55 µg/l) and for BW 284 C51 of 10 nM (= 5.7 µg/l) are acceptable, whereas the detection limit of 1 µM (= 0.26 mg/l) for pyridostigmine is not. However, the sensitivity of the method can be increased. It was shown that the optimal temperature for the inhibition of acetylcholine esterase with

paraoxon is about 30 °C [17]. All measurements presented here were performed at room temperature. Not only the temperature, but also the flow-rate could be increased to enhance the sensitivity of the method towards the inhibitors (Figs. 11 & 15). However, this would lead to an additional consumption of enzyme substrate and a shorter lifetime of the immobilized enzyme. The mixing ratio of substrate:buffer:inhibitor solutions could also be changed in favour of the inhibitor solution. It is also known, that choline esterase enzymes from different sources show differing sensitivities towards the various inhibitors [20]. For the detection of a certain pesticide it would be possible to choose another more appropriate enzyme. Finally the signal/noise ratio of the measuring system can be improved through optical and electronical refinements. With such an optimized system, very small signal changes can be recognized and the detection limits could be lowered by one or two orders of magnitude.

For the determination of BW 284 C51 and pyridostigmine, no data were available from the literature. The determination of paraoxon has been described several times. In a radiometric method with an incubation time of 60 minutes and a total time requirement of at least two hours the detection limit was as low as 4.29×10^{-10} M paraoxon [11]. In a colorimetric method the incubation time was two hours and the whole procedure lastet ca. 2.5 hours [11]. A very tedious procedure with an immunoassay coupled to an enzymatic reaction (total time consumption ca. 2.5 hours) resulted in a detection limit of 1 nM paraoxon [11].

The method presented here doese not require radioactivity or antigenes. A detection limit of 200 µM paraoxon after a measuring time of 21 minutes seems to be very acceptable especially in view of the possible enhancement through opto-electronics. It would be no problem to measure the signal changes caused by the inhibition process after one hour and to increase the sensitivity. For practical purposes, a method which gives a signal change after one hour is still more useful than a method which requires two hours, especially in a pesticide alarm system.

In a flow injection analysis system (FIA) a rapid and somewhat sensitive determination of paraoxon was possible [17]. Using this method, 60 samples per hour with a detection limit of 400 nM paraoxon or 30 samples per hour (in the stopped-flow mode) with a detection limit of 8 nM were measured. Both, the inhibitor and the substrate solutions, were injected into a buffer carrier stream. The synthetic substrate was hydrolyzed in an enzyme reactor and the product reacted with a second reagent to form a dye whose absorption was measured at 500 nm. According to the authors, the rapid determination of the pesticide was possibl because inhibitor and substrate reached the enzyme at the same time and a competition in binding to the active site occured. No long incubation times were necessary. This method seems to be applicable to our system and will be investigated in the near future. A rapid analysis of small sample volumes is advantageous in many cases, although a more complex and expensive instrumentation is required.

The method proposed in this work could be used for continuously monitoring a drinking or ground water reservoir. The signal change produced for example by an organophosporus or carbamate pesticide may serve as an alarm to switch of the water supply or to start more detailed investigations, such as qualitative and quantitative analysis or search for the origins of the pesticides. It would also be of interest to test the response of the system towards the so-called nerve gases, since these substances are closely related to the organophosphorus pesticides. It is known that these chemical warfare agents are very potent inhibitors of the enzymes acetyl- and butyrylcholine esterase. Therefore these compounds must be detectable with our system. The identification of nerve gases over large distances could provide a simple and sensitive alarm system, not only in cases of war but also in case of an accident in the production, transport or destruction of such substances.

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Figures

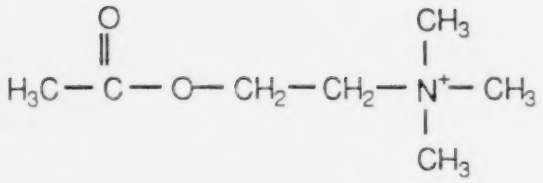

Fig. 1. Acetylcholine

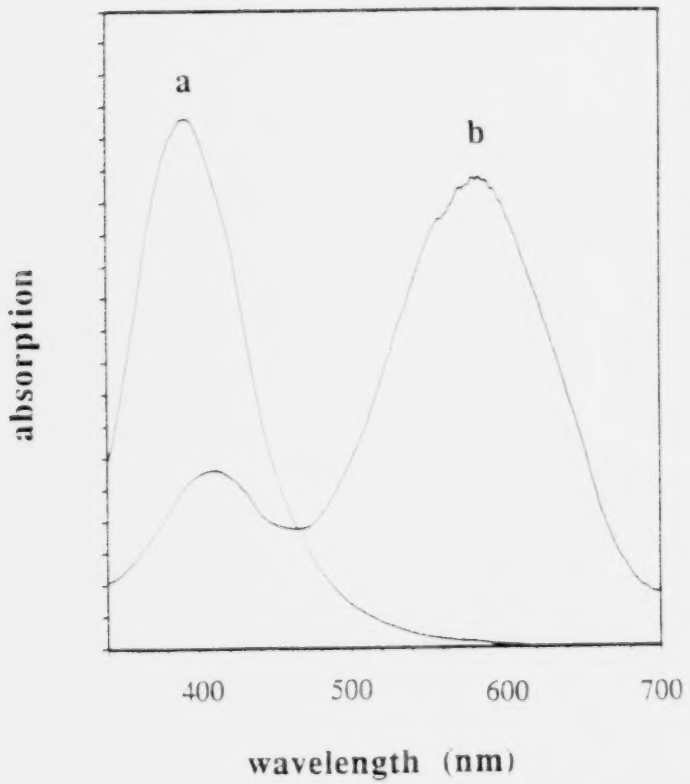

Fig. 2. Absorption spectra of Substrate A (a) and its product of hydrolysis (b) at pH 7.5

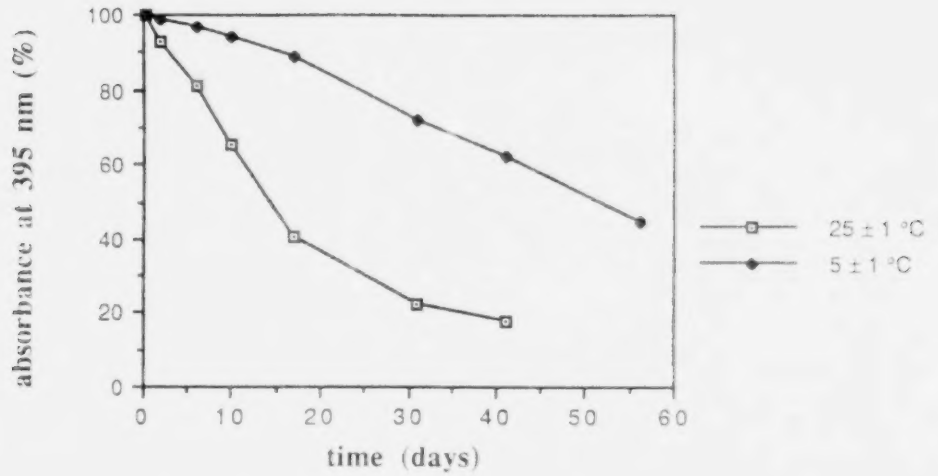

Fig. 3. Stability of Substrate A in destilled water at 25 ± 1 °C and at 5 ± 1 °C in the dark

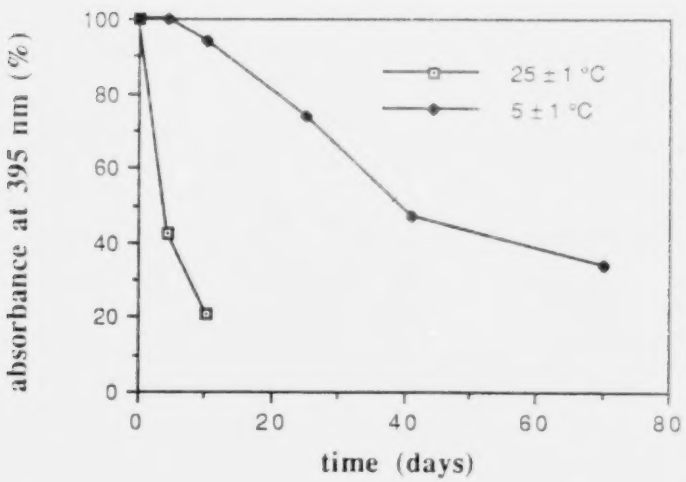

Fig. 4. Stability of Substrate B in destilled water at 25 ± 1 °C and at 5 ± 1 °C in the dark

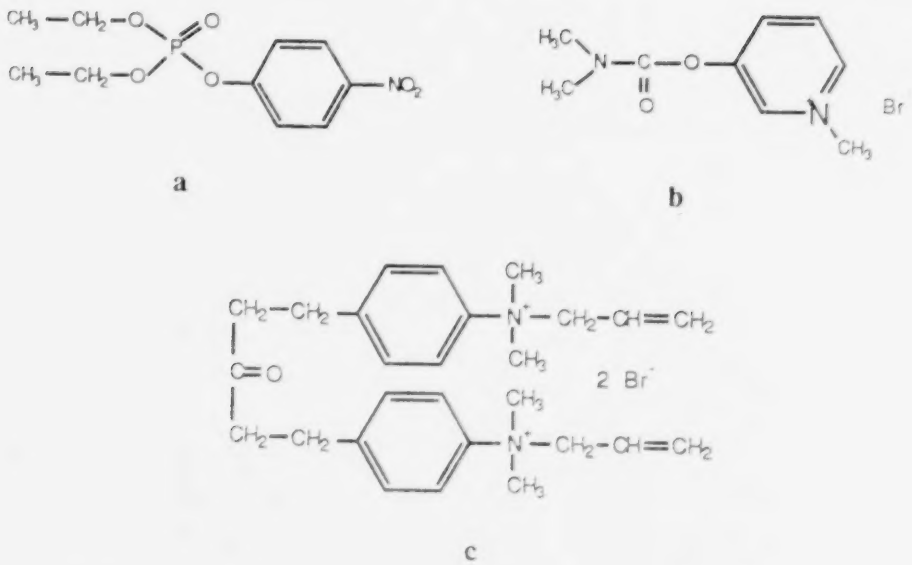

Fig. 5. (a) Paraoxon; (b) pyridostigmine bromide; (c) BW 284 C51

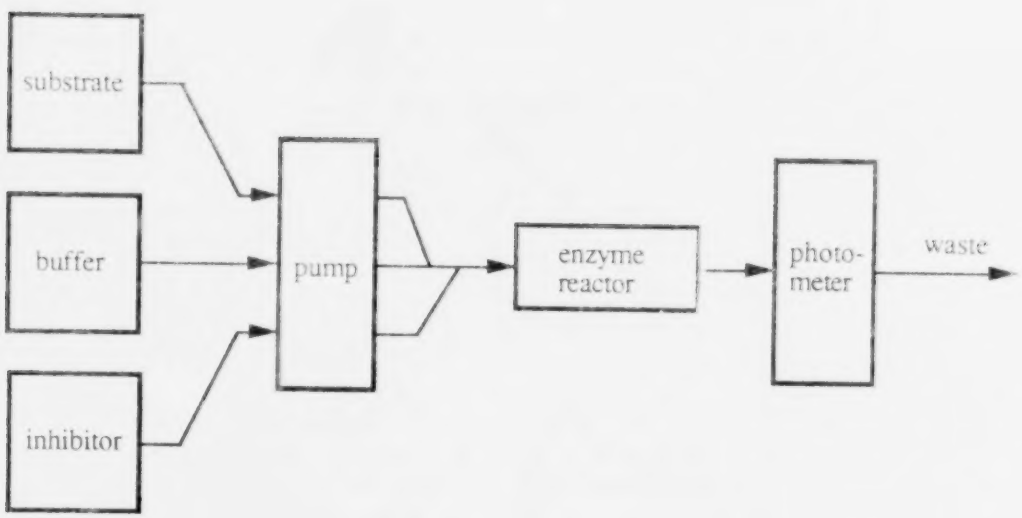

Fig. 6. Experimental set-up for measurements with an UV VIS-spectrophotometer

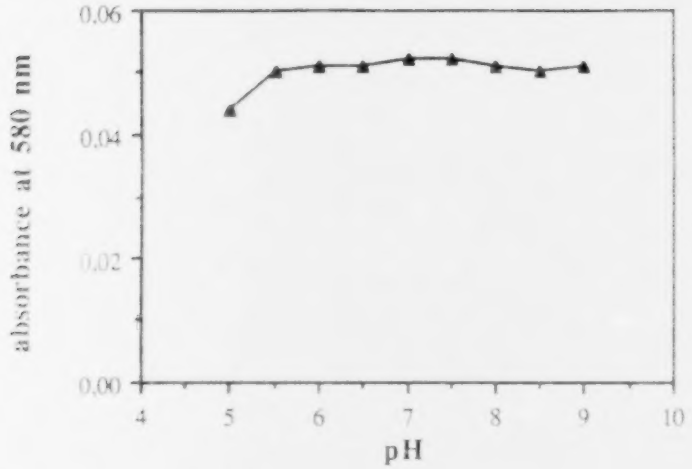

Fig. 7. pH-dependence of the enzymatic hydrolysis of Substrate A at a flow-rate of 0.58 ml/min.

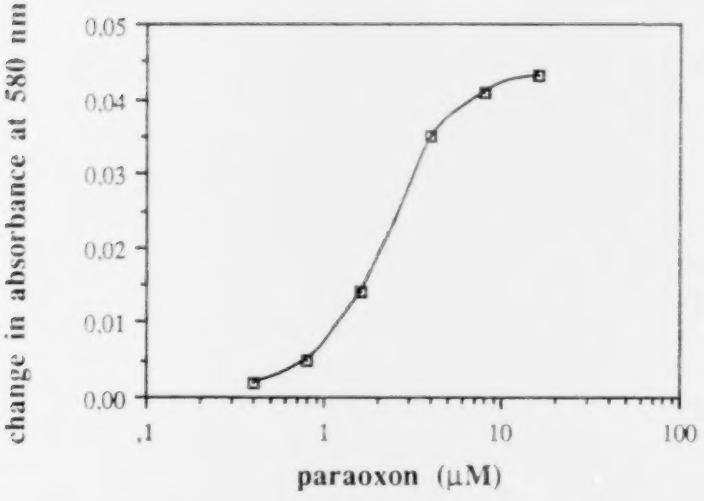

Fig. 8. Inhibition of acetylcholine esterase with paraoxon - calibration graph (signal changes after 21 min; pH 7.5; flow-rate: 1.8 ml/min)

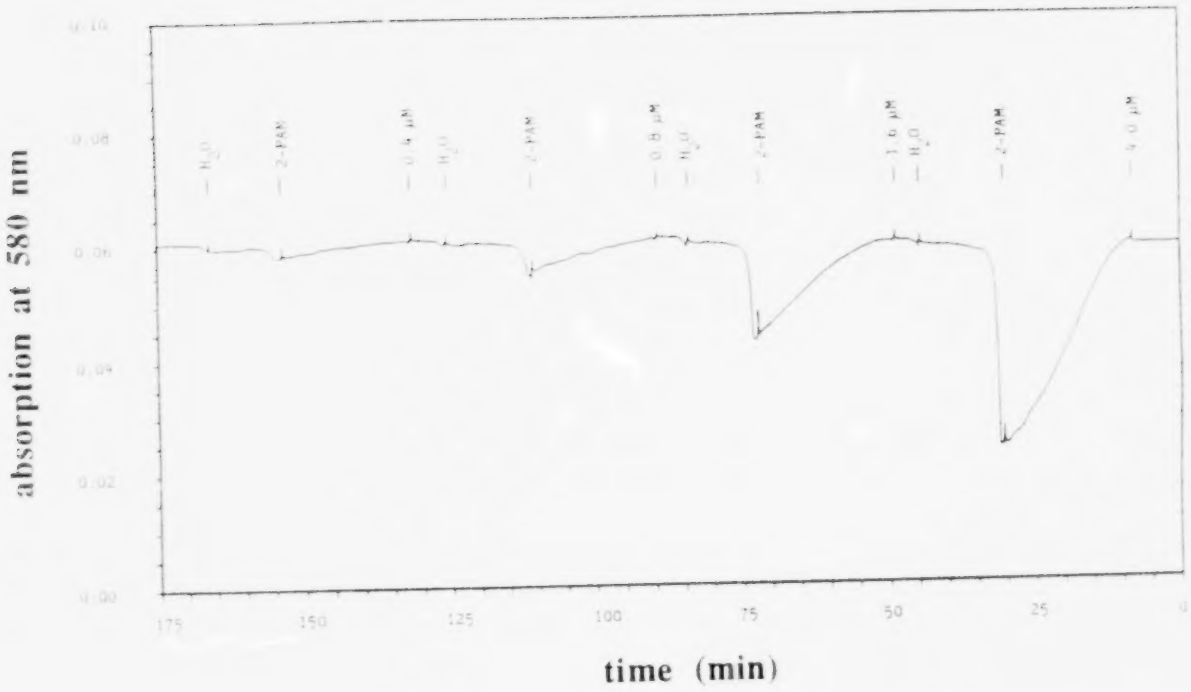

Fig. 9. Inhibition of acetylcholine esterase with paraoxon - time dependence (pH 7.5; flow-rate: 1.8 ml/min)

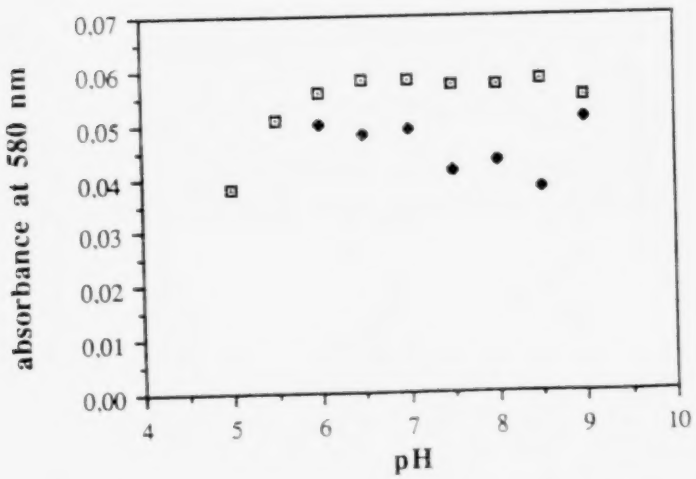

Fig. 10. Inhibition of acetylcholine esterase with paraoxon - pH-dependence (signal changes after 21 min; flow-rate: 1.46 ml/min)

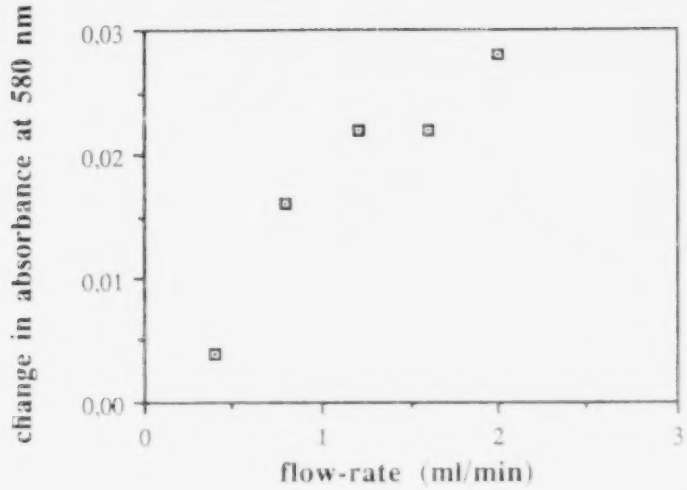

Fig. 11. Inhibition of acetylcholine esterase with paraoxon - flow-rate dependence (signal changes after 21 min; pH 7.5)

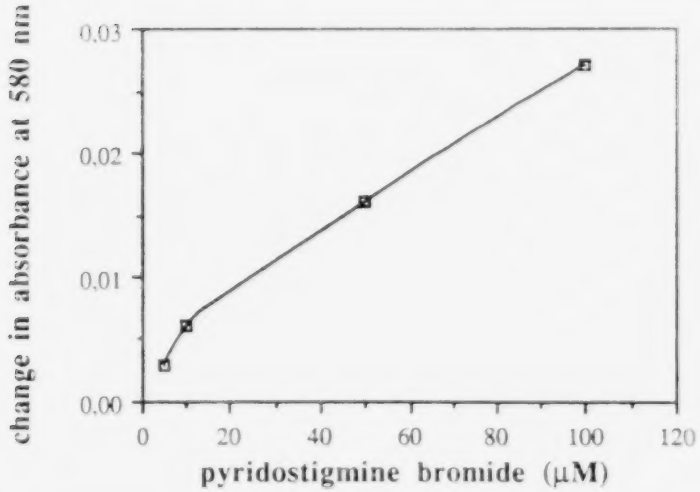

Fig. 12. Inhibition of acetylcholine esterase with pyridostigmine - calibration graph (signal changes after 21 min; pH 7.5; flow-rate: 1.8 ml/min)

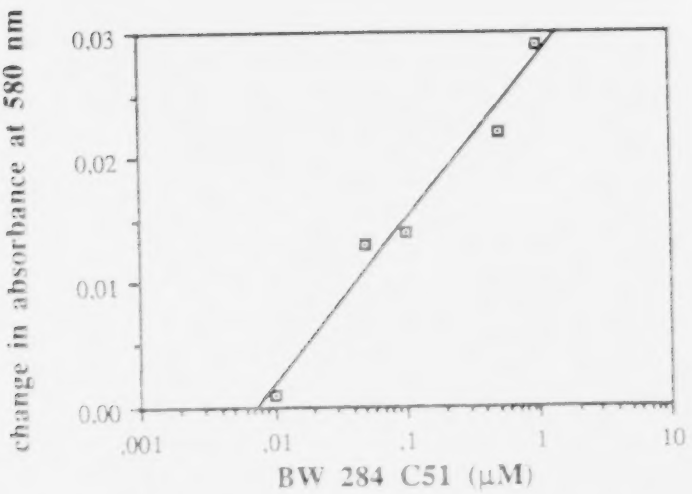

Fig. 13. Inhibition of acetylcholine esterase with BW 284 C51 - calibration graph (steady-state response; pH 8.0; flow-rate: 1.2 ml/min)

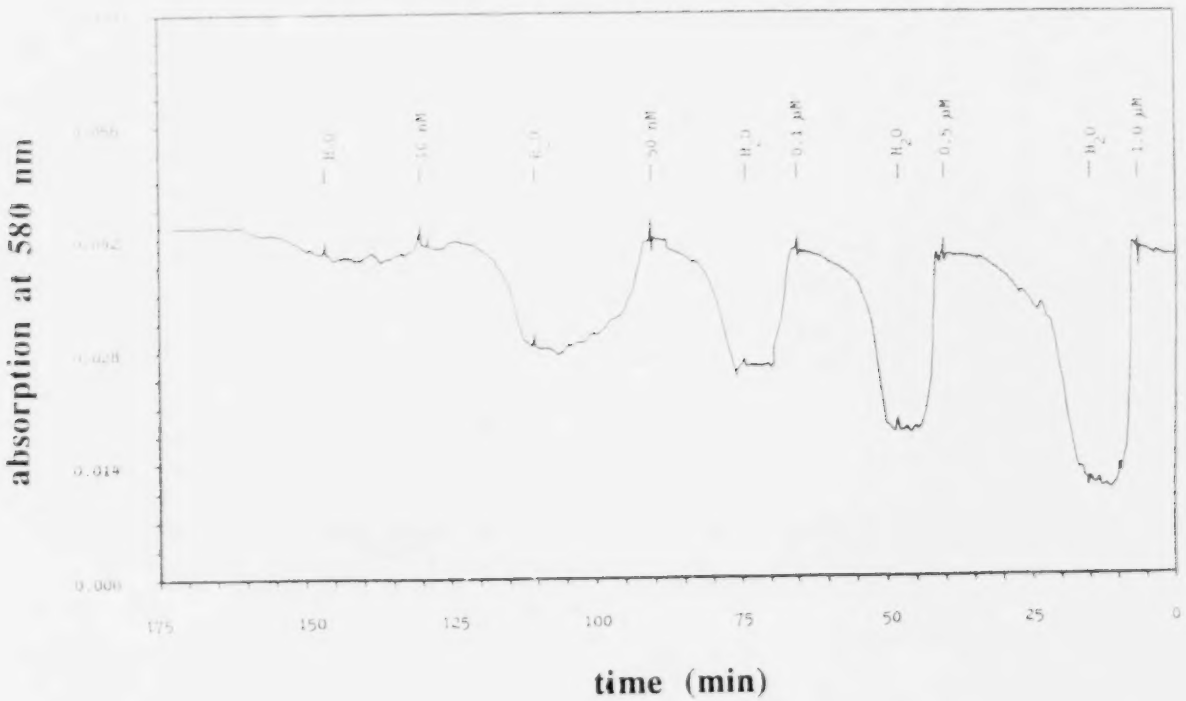

Fig. 14. Inhibition of acetylcholine esterase with BW 284 C51 - time dependence (pH 8.0; flow-rate: 1.2 ml/min)

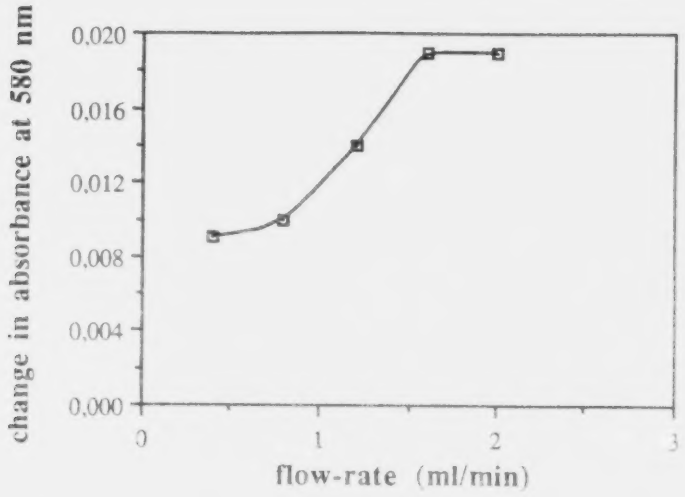

Fig. 15. Inhibition of acetylcholine esterase with 0.1 µM BW 284 C51 - flow-rate dependence (steady-state response; pH 8.0; flow-rate: 1.2 ml/min)

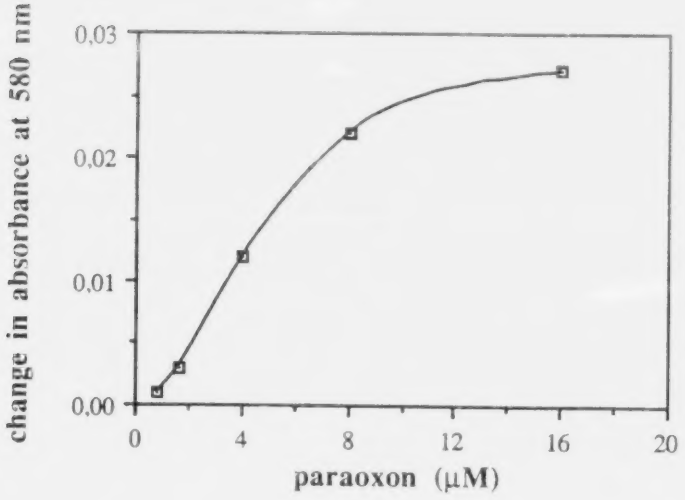

Fig. 16. Inhibition of acetylcholine esterase with paraoxon - calibration graph (signal changes after 21 min; pH 7.5; flow-rate: 1.8 ml/min)

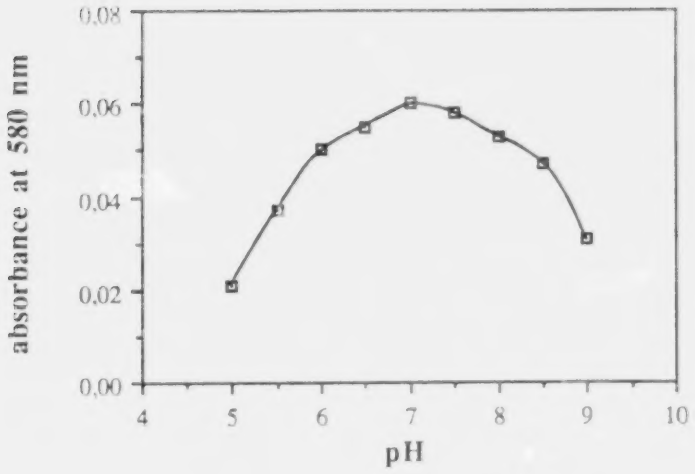

Fig. 17. pH-dependence of the hydrolysis of Substrate B by butyrylcholine esterase immobilized on aminopropyl-CPG with glutaraldehyde (flow-rate: 1.8 ml/min)

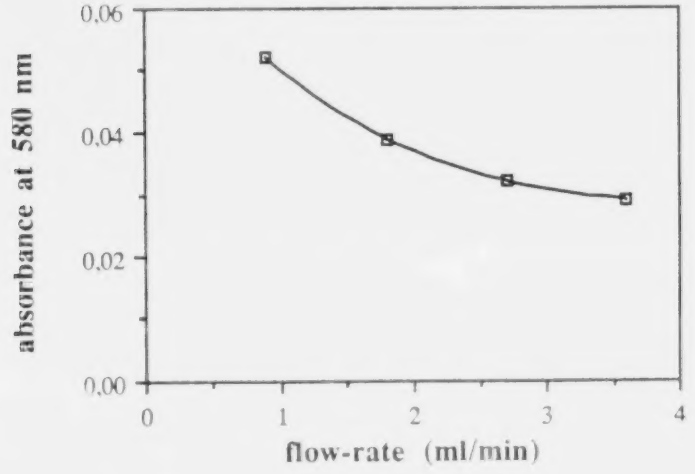

Fig. 18. Flow-rate dependence of the hydrolysis of Substrate B by butyrylcholine esterase immobilized on aminopropyl-CPG with glutaraldehyde (pH 7.5)

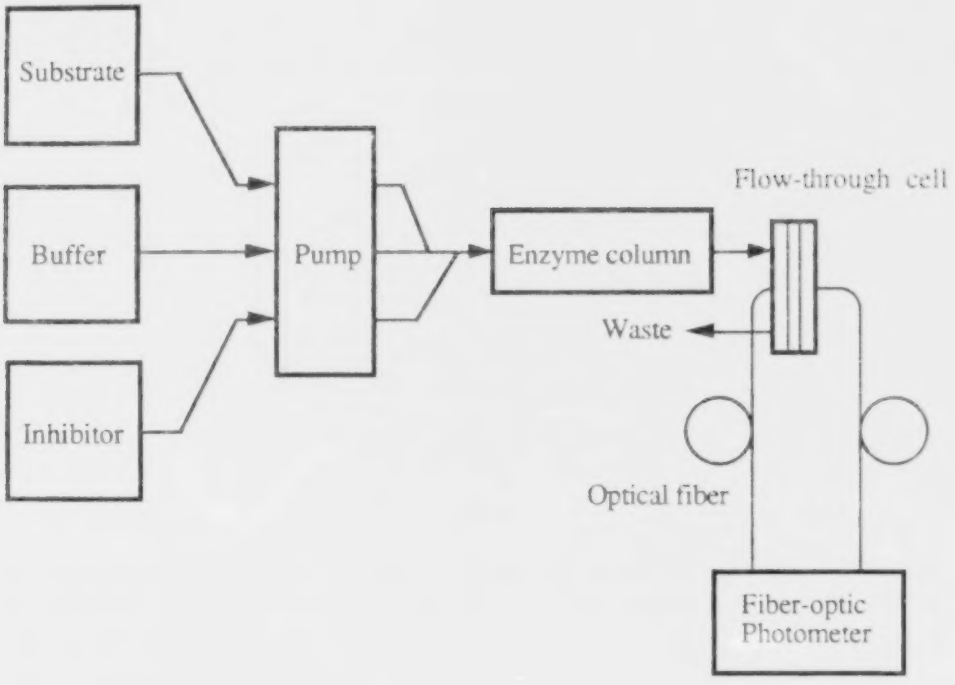

Fig. 19. Experimental set-up for measurements with a fiber-optic photometer

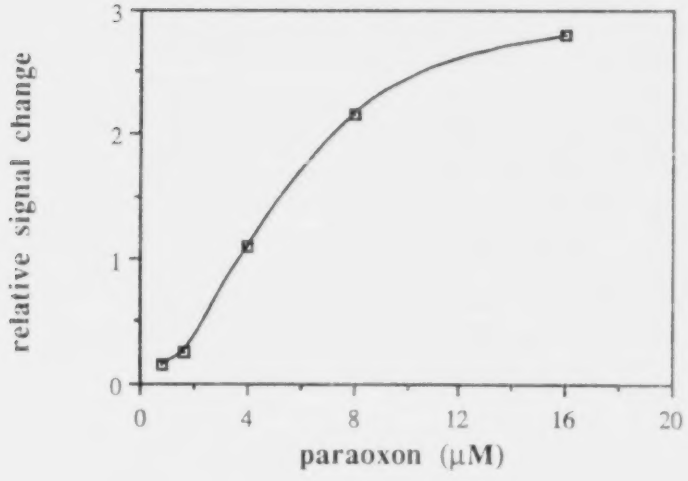

Fig. 20. Inhibition of acetylcholine esterase immobilized on VA-Epoxy-Biosynth with paraoxon (signal changes after 21 min; pH 7.5; flow-rate: 1.8 ml/min)

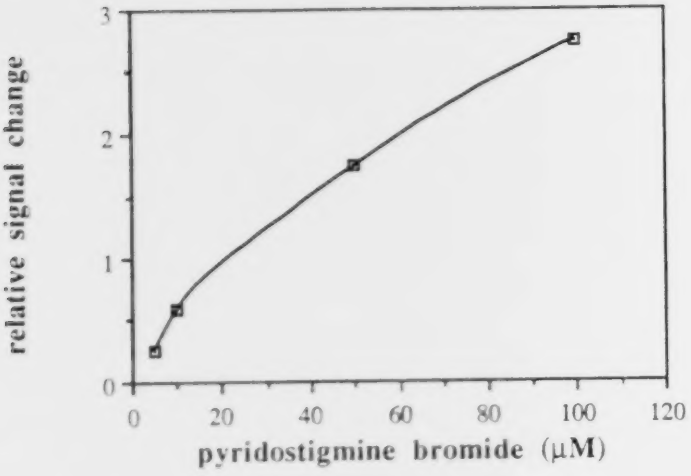

Fig. 21. Inhibition of acetylcholine esterase immobilized on aminopropyl-CPG with pyridostigmine (signal changes after 21 min; pH 7.5; flow-rate: 1.8 ml/min)

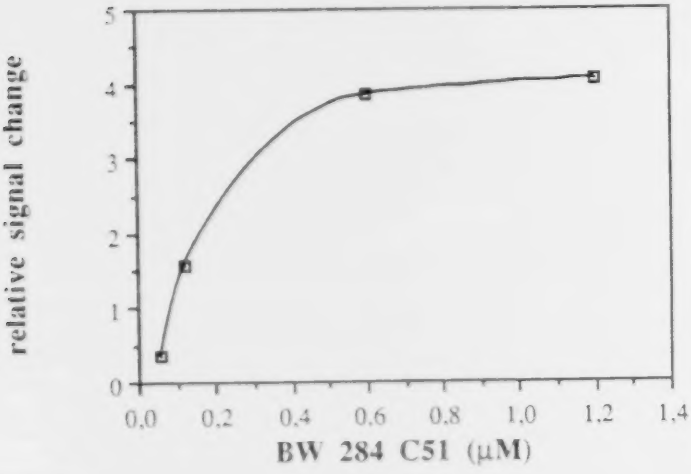

Fig. 22. Inhibition of acetylcholine esterase immobilized on aminopropyl-CPG with BW 284 C51 (steady-state response; pH 7.5; flow-rate: 1.8 ml/min)

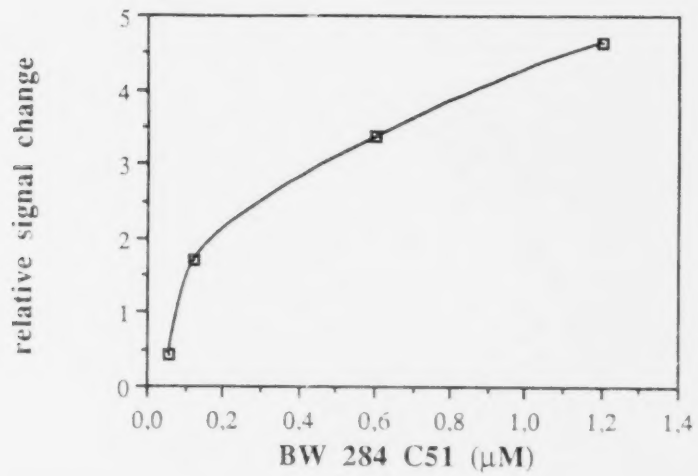

Fig. 23. Inhibition of acetylcholine esterase immobilized on aminopropyl-CPG with BW 284 C51 (steady-state response; pH 7.5; flow-rate: 1.8 ml/min)

CONFERENCE ON DISARMAMENT

CD/1077
23 May 1991

Original: ENGLISH

LETTER DATED 22 MAY 1991 FROM THE ACTING REPRESENTATIVE OF THE UNITED STATES OF AMERICA ADDRESSED TO THE PRESIDENT OF THE CONFERENCE ON DISARMAMENT TRANSMITTING A STATEMENT ISSUED BY THE PRESIDENT OF THE UNITED STATES OF AMERICA CONCERNING THE UNITED STATES INITIATIVE FOR COMPLETING THE NEGOTIATIONS ON A CHEMICAL WEAPONS CONVENTION, AND A WHITE HOUSE FACT SHEET ON THE INITIATIVE

I have the honour to forward to you the attached statement issued by the President of the United States, Mr. George Bush, concerning the United States initiative for completing the negotiations on a Chemical Weapons Convention, and a White House Fact Sheet on the initiative.

I ask that you take the appropriate steps to enter this statement and fact sheet as official documents of the Conference on Disarmament and to have them distributed to all member delegations and non-member States participating in the work of the Conference.

(Signed) M. LYALL BRECKON
Acting Representative of the
United States of America to
the Conference on Disarmament

THE WHITE HOUSE

Office of the Press Secretary

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13 May 1991

STATEMENT BY THE PRESIDENT ON CHEMICAL WEAPONS INITIATIVE

The Gulf war has once again raised the spectre of chemical weapons and demonstrated that unscrupulous régimes can and will threaten innocent populations with these weapons of terror so long as we permit them to exist. These stark events renew and reinforce my conviction, shared by responsible leaders around the world, that chemical weapons must be banned - everywhere in the world.

The world's best hope for this goal is the Chemical Weapons Convention now being negotiated by the Conference on Disarmament in Geneva. The United States is committed to the early, successful completion of this Convention and I am today announcing a number of steps we will take to accelerate the negotiations and achieve an effective chemical weapons ban as soon as possible. I hope this initiative also will spur other nations to commit themselves equally to this critical objective.

I have instructed the United States negotiating team in Geneva to implement my decisions at the next session which begins 14 May. To demonstrate the United States commitment to banning chemical weapons, we are formally foreswearing the use of chemical weapons for any reason, including retaliation, against any State, effective when the Convention enters into force, and will propose that all States follow suit. Further, the United States unconditionally commits itself to the destruction of all our stocks of chemical weapons within 10 years of entry into force and will propose that all other States do likewise. We will offer technical assistance to others to do this efficiently and safely.

In addition, we will call for setting a target date to conclude the Convention and recommend the Conference stay in continuous session if necessary to meet the target. The United States also will propose new and effective verification measures for inspecting sites suspected of producing or storing chemical weapons. To provide tangible benefits for those States that join the Convention and significant penalties for those that fail to support it, the United States will propose the Convention require parties to refuse to trade in chemical weapon-related materials with States that do not join in the Convention. The United States reaffirms that we will impose all appropriate sanctions in response to violations of the Convention, especially the use of chemical weapons.

These steps can move the world significantly closer to the goal of a world free of chemical weapons. I call upon all other nations to join us in the serious and cooperative pursuit of this important goal.

THE WHITE HOUSE

Office of the Press Secretary

FACT SHEET ON CHEMICAL WEAPONS INITIATIVE

Noting that the recent conflict in the Persian Gulf had brought into play once again the spectre of the use of chemical weapons against innocent populations, the President announced a new series of steps in his long-term goals of ridding the world of these weapons of terror. The President reiterated his belief that the best hope for achieving that goal is the Chemical Weapons Convention (CWC) - a multilateral treaty that would completely ban the production, possession, transfer and use of chemical weapons - under negotiation in the Conference on Disarmament (CD) in Geneva.

The primary purposes of this initiative are to strengthen the prospects of an early successful conclusion of the Convention, to demonstrate the commitment of the United States to achieving an effective ban on CW as soon as possible, and to provide incentives to other nations to commit themselves equally to this critical objective. To this end, the President declared that the United States delegation to the CD would take the following actions when the next session of the talks on the CWC resumes 14 May 1991:

1. The United States will call for the resolution of all major outstanding issues in the CD talks in Geneva on the CWC by the end of 1991 and the completion of the CWC within twelve (12) months.

2. To help reach these goals and to ensure that no time is lost and no opportunity is wasted to bring the negotiations to a successful conclusion soon, the United States will propose that the CW ad hoc negotiating committee of the CD stay in continuous session in Geneva, beginning with the May session. The United States is prepared to keep its ambassador at the table and to reinforce its delegation as necessary to support this schedule.

3. The United States will formally declare its intention to become an original party to the CWC and will call upon all States publicly to commit to becoming original parties to the treaty and to declare their CW stocks.

4. The United States will drop its position that States retain a right of retaliation with CW as long as they have CW stocks if CW are used against them first. The United States will formally foreswear the use of CW for any reason, including retaliation in kind with CW, against any State, effective when the CWC enters into force. The United States will also propose a provision for the CWC prohibiting the use of CW for any reason.

5. The United States will drop its position that we must be allowed to keep 2 per cent of our CW stockpile (500 tons) until all CW-capable States have joined the Convention. The United States will unconditionally commit itself to the destruction of all of its CW stocks and former production facilities within 10 years of entry into force of the CWC. We will call upon all States to follow our lead and to begin the destruction of their CW stocks and production facilities as soon as possible. We will propose provisions for the

CWC that require all parties to begin the unconditional destruction of their CW stocks and production facilities as soon as the CWC enters into force, and to complete their elimination within 10 years of that date.

6. The United States will provide practical and effective help to other States to assist in the speedy, safe and environmentally sound destruction of CW wherever they may be found. We will:

- Create a Task Force comprised of United States Government and private industry representatives to develop proposals on how the United States can advise and assist States in CW destruction.

- Provide technical destruction assistance (e.g., visits to United States destruction facilities, blueprints and technical advice) to States parties that have CW located on their territory.

- Propose the creation of a CD Working Group on CW destruction and offer to provide United States experts to it.

7. The United States will provide extensive information to other States on United States export controls on CW-related materials and technology and our domestic legislation to enforce them. We will call on all States to enact similar controls, thereby enhancing the control of CW proliferation as we work toward a complete and formal ban.

8. Aside from the significant intrinsic benefits that will flow to all parties from a total ban on CW, there should be tangible benefits for those States that join the Convention and enhance its strength, and specific penalties for those States that fail to support it in the most fundamental and important way by not joining it. Therefore, the United States will propose provisions for the CWC that require parties to refuse to trade in CW-related materials with States that do not become parties within a reasonable time after entry into force.

9. The United States will reaffirm our position that there should be sanctions against those States that violate the CWC and especially severe sanctions against any State that initiates the use of CW. The United States declares that it will consider violations of the CWC, especially the initiation of the use of CW, as extremely grave breaches of international law, and that the United States will impose all appropriate sanctions.

10. The United States will reaffirm our commitment to a strong and forthright verification régime. We will offer additional concrete proposals to address the central issue of inspecting suspect sites. We will call on the CD to adopt our position for challenge inspections of such sites.

CONFERENCE ON DISARMAMENT

CD/1078
CD/CW/WP.340
30 May 1991

Original: ENGLISH

LETTER DATED 30 MAY 1991 FROM THE DEPUTY PERMANENT REPRESENTATIVE OF NORWAY TO THE DEPUTY SECRETARY-GENERAL OF THE CONFERENCE ON DISARMAMENT TRANSMITTING A DOCUMENT ENTITLED, "VERIFICATION OF ALLEGED USE OF CHEMICAL WARFARE AGENTS: APPLICATION OF PROCEDURES AFTER A SIMULATED CHEMICAL ATTACK ON AN AIR BASE"

I have the honour to enclose a document entitled, "Verification of alleged use of chemical warfare agents: application of procedures after a simulated chemical attack on an air base".

I would be grateful if this document could be issued as an official document of the Conference on Disarmament as well as in the series of Working Papers for the Ad Hoc Committee on Chemical Weapons.

(Signed) Torbjørn Aalbu
Minister
Deputy Permanent Representative
to the Conference on Disarmament

GE.91-61020

VERIFICATION OF ALLEGED USE OF CHEMICAL WARFARE AGENTS

APPLICATION OF PROCEDURES AFTER A SIMULATED CHEMICAL ATTACK ON AN AIR BASE

Introduction

An extensive study of the possibility of verifying alleged use of chemical weapons was initiated by the Norwegian Ministry of Foreign Affairs in 1981. The research has been carried out by the Division for Environmental Toxicology of the Norwegian Defence Research Establishment at Kjeller near Oslo. The annual reports and working papers submitted to the Conference on Disarmament have been compiled in the publication Contributions by Norway to the conference on Disarmament 1982-1987 (document CD/813 dated 7 March 1988).

The 1988 report was mainly concerned with the development of complete procedures for verification of alleged use of chemical weapons, (CD/857 of 12 August 1988 and CD/861 of 22 August 1988), while the research in 1989 focussed on the development and testing of headspace gas chromatography, a new supplementary technique (CD/936 of 21 July 1989 and CD/940 of 31 July 1989).

The sample preparation method developed by the Norwegian research programme is based on the adsorption of chemical warfare agents from aqueous solutions onto a polymer sorbent and their subsequent removal by an organic solvent. This technique, referred to as sorbent extraction, was optimized in the 1989-1990 programme in order to isolate as efficiently as possible chemical warfare agents and related compounds from different sample materials. The procedure has been made as general as possible for use in screening samples suspected of being contaminated with chemical warfare agents. These results and conclusions were summarized in document CD/1008 dated 26 June 1990, and the research report was presented to the Conference with document CD/1019 dated 23 July 1990.

Based on the work carried out during the Norwegian research programme, a complete procedure for the verification of alleged use of chemical weapons has been developed, including the construction of a mobile field laboratory. This laboratory, contained in a wooden case, holds all the equipment and chemicals needed to carry out sampling, sample preparation and preliminary analyses in the field. The most important parts of this verification procedure are location of the contaminated area, sample collection, field analysis, sample transportation, sample preparation and sample analysis.

When the Chemical Weapons Convention enters into force, the Technical Secretariat will receive the results from the laboratories which analyze samples taken during the inspection of an area where use of chemical weapons is alleged to have taken place. To allow correct conclusions to be drawn from the results provided, it is important that the history of the

samples has been properly recorded. All available information from the inspection of the contaminated area, sampling, sample preparation and transport should be recorded and passed on to the Secretariat. Most of this information should also be passed on to the analytical laboratories, to make it easier to select the best suited analytical method and to evaluate the reliability of the results. It is also important that the samples are sealed and secured to prevent tampering during transport.

During this research programme, the evaluation of verification procedures by field exercises has been emphasized, in order to reveal problems that would not occur during laboratory testing.

This year's report is divided into two parts. The main part is an application of the complete procedures for verification of alleged use of chemical weapons, developed during the Norwegian research programme. It deals with handling of samples from a simulated attack with chemical weapons on an air base. Samples were collected in the area in question and treated in accordance with the procedures. In addition to the samples collected after the simulated attack, some samples of soil, sand, water and protective clothing were spiked with chemical warfare agents in the laboratory, transported to the air base, where they were exposed to the prevailing weather, and then treated in the same manner as the samples collected.

A form has been drawn up on which the sampling team should enter all information acquired during the localization of the contaminated area, sample collection, sample preparation, field analyses and sample transport. A transport log has also been drawn up which would be filled in by the personnel in charge of each section of the transport chain in order to keep a complete record of the history of the samples.

The second part of the report describes further improvement of the procedures for handling samples. This includes testing methods for homogenization of samples, and a comprehensive testing of solvents for extraction of different chemical warfare agents from various sample materials.

The report confirms earlier results on the advantages of the sorbent extraction technique for the detection of chemical warfare agents, and describes further improvements of the sample preparation and handling procedures. The report is a concrete contribution by experts at the Norwegian Defence Research Establishment to the negotiations on a Chemical Weapons Convention closely related to the Protocol on Inspection Procedures (CD/1046, Appendix I, Addendum, Part IV: Procedures in cases of alleged use of Chemical Weapons) and the relevant General Assembly resolutions on measures to uphold the authorities of the 1925 Geneva Protocol, in particular resolution 45/57 C endorsing the proposals of the group of qualified experts concerning technical guidelines and procedures to guide the Secretary-General in the conduct of

timely and efficient investigation of the reports of use of chemical and bacteriological (biological) or toxin weapons (document A/44/561 dated 4 October 1989).

Field exercise

An attack on an air base with chemical warfare agents was simulated by exploding a bottle filled with 200 g of the nerve agent simulant dipropyleneglycol monomethyl ether (DPM) 1.5 m above the ground. After the explosion, all visual evidence was removed before a team from the Norwegian Defence Research Establishment was called upon to verify the alleged use of chemical warfare agents on the air base. This team was unaware of the exact site of the explosion, and was required to identify the contaminated area, carry out field analyses, collect samples and treat them according to the Norwegian procedure, and transport them to the laboratory.

Five hours after the explosion of the simulant the inspection team arrived at the air base, and was transported to the area where eyewitnesses had seen a suspicious explosion of an artillery shell. The inspection was carried out as it would have been after a real incident, and the inspectors used respirators and complete protective clothing throughout the localization and sample collection phases. An area of 100 m x 100 m pointed out by the eyewitnesses was first inspected visually by two members of the team in order to find any remnants from the explosion. This inspection took about 7 minutes, but gave no indication of where the explosion had taken place. The inspection team then used a chemical agent monitor (CAM) and detection paper to localize the possible contaminated area. After 20-25 minutes total search time, an area contaminated with a nerve agent simulant was located; another 10 minutes were used to define the extent of this area. Detection paper was used to confirm the observations made with the CAM. The contaminated area was found to be 15m x 10m.

Twenty samples were collected throughout this contaminated area. Most of the samples were, however, collected near the point which on the CAM showed the highest concentration of the DPM simulant. Samples of about 50 g of soil, sand or water were collected. Reference samples from outside the contaminated area were also collected. The samples were placed in glass bottles with airtight screw caps, coded by sample number and sealed with wax, and then transported back to the laboratory. No further analyses were carried out on these samples.

It is extremely important to record information on the samples collected, the sampling methods and sampling site to allow correct conclusions to be drawn from the results obtained by sample analysis. A three-page form has therefore been drawn up

for use by the sampling team during the inspection. This form is included as an annex to this paper. On the first page, information on the sampling should be entered, including the reason for sampling, an indication of the priority or importance of the samples, description of samples and sampling site, sampling methods and results of field analyses. On the second page, a sketch of the sampling site should be drawn, indicating topography, any bomb craters observed and wind direction. The positions of the sampling sites should also be indicated on this sketch. The third page should be filled in if information on the attack is available. Reports from eyewitnesses, casualties, the effect on vegetation and the weather conditions since the attack should be entered here.

In addition to samples from this simulated attack, samples of 50 g of soil, sand and water, and 0.05 g (20 cm^2) of protective clothing were spiked in the laboratory with 1 mg of each of the nerve agents GA, GB, VX, and the vesicants Lewisite (L) and mustard (H). These samples were placed in closed vessels and transported to the air base where the vessels were opened and the samples exposed to the prevailing weather for 20 hours. Some of these samples were then treated in the field according to the standard Norwegian procedure and the others transported back to the laboratory and used for evaluation of different sample preparation methods.

In order to have a first indication of which chemical was present, some of the samples spiked with real chemical warfare agents were analyzed in the field using thin layer chromatography (TLC). The agents GA, GB, VX and H were detected in all sample materials (soil, sand, water and protective clothing). As Lewisite is rapidly hydrolyzed in water, it could not be detected in the aqueous samples.

After exposure to the prevailing weather for 20 hours, three replicates of each of the spiked samples were washed with 50 ml water and passed through a combination of C_{18} cartridges (top) and NH_2 cartridges (bottom) using a hand operated vacuum system. The cartridges were pre-wetted with 1 ml methanol and 10 ml water before application of the samples. The cartridges were labelled, put in a sealed plastic bag and placed in a thermos flask of dry ice for transport to the laboratory. The rest of the spiked samples were transported untreated in closed containers back to the laboratory for further analyses; a military aircraft was used for the transport, and military regulations were followed. All relevant information on the samples and sample preparation was entered on the form previously described. One copy of the form was transported together with the samples, and one copy was retained by the sampling team. A transport log has further been drawn up to follow the samples during transportation. One log followed each parcel, and the date and time were entered by the personnel in charge of each stage of transportation from the sampling site to the destination. The maximum permissible temperature during transport was filled in by the sampling team for the benefit of the personnel in charge of transport.

A transport log is shown as an annex to this paper.

At the laboratory, the chemical compounds were eluted from the C_{18} and NH_2 cartridges and the eluates analyzed by gas chromatography. One portion of each of the untreated samples was also analyzed by headspace gas chromatography without pretreatment.

The results of the analyses of the spiked and weathered samples show that GA, GB, VX and H were found in all sample materials using the C_{18} sorbent extraction technique. The headspace method is not useful for low-volatile compounds like VX, but detected all the compounds except sarin (GB) in sand and tabun (GA) and mustard (H) in soil samples. Because lewisite (L) decomposes very rapidly in contact with water, this compound was not detected in any of the samples analyzed.

Laboratory experiments

When the samples are received at the laboratory after transport from the field, they are generally split into several sub-samples for analysis by several different methods. To be sure that the sub-samples contain equal amounts of the chemical compounds to be determined, it is important that the main samples are homogenized and split properly. A method of homogenization of solid samples before splitting into sub-samples has been developed using a shaking machine for dry samples and a mortar for wet samples. In these experiments, the sarin impurity diisopropyl methylphosphonate (DIPMP) was used as a model substance and added to sand, soil and glass spheres at several different concentrations. When the homogenized samples were split into ten equal sub-samples and analyzed, DIPMP was found in all samples with satisfactory standard deviations.

In addition, the organic solvents methanol, acetone, dichloromethane, tetrachloromethane, chloroform, isopropanol, diethyl ether, ethylacetate, hexane and the mixtures methanol:chloroform (1:1) and chloroform:diethyl ether (1:1) were tested to evaluate which one most efficiently extracts selected chemical warfare agents and related compounds from different sample materials. The chemical compounds tabun (GA), sarin (GB), soman (GD), VX, mustard gas (H), and methyl 1,2,2-trimethylpropyl methylphosphonate (MTMP) were tested on the materials sand, soil, paper and butyl rubber. This experiment showed that the highest recovery rates were obtained with the methanol/chloroform mixture. This mixture also showed the smallest variations (SD) among the sample materials. There were only small differences between the results obtained with other extraction solvents, except for hexane, which consistently gave the lowest extraction yields. One problem observed with the methanol/chloroform mixture was that phase separation occurred with some types of samples.

Conclusions from field experiment

To evaluate the complete verification procedure, including the usefulness of the sampling forms and transport log, an attack with chemical weapons on an air base was simulated. The sampling team used the chemical agent monitor (CAM) and detection paper to localize and define the contaminated area in about 30 minutes. The CAM proved especially useful for this purpose.

To allow correct conclusions to be drawn from the results of the sample analysis, it is extremely important to record information during location of a contaminated area, sample collection, sample preparation, field analysis and sample transport . A form has therefore been drawn up for use by the sampling team during the inspection. This should be completed with information about the reason for taking samples, a description of the sample site, sampling methods, weather conditions and results of the field analysis. If eyewitness accounts or other information on the chemical attack is available, this should be included, together with reports on the inspection of any casualties. One copy of the form should be sent to the laboratory together with the samples and one copy retained by the sampling team. A transport log has also been drawn up, and this should follow each parcel during transportation. The date and time should be entered in this log by personnel in charge of each section of the transport.

The field exercise showed that thin layer chromatography (TLC) can be used as a rapid method for screening samples of chemical warfare agents in the field close to the contaminated area. This method gives an initial indication of which chemical agent is present in the samples, and could make it easier to select the methods of sample preparation and analysis which give the most reliable results in the laboratory.

The analysis of the samples spiked with real chemical warfare agents showed that the sorbent extraction (C_{18}) method could detect all spiked agents in all sample materials. The greatest advantage of this method is that chemical warfare agents can be isolated from the sample materials in the field immediately after sample collection. This minimizes the loss of information due to decomposition and evaporation of the agents during transport to the laboratory. In addition, the sorbent extraction technique is easy to carry out and does not require large amounts of equipment and chemicals.

Conclusions from Laboratory experiments

It is important that the samples are properly homogenized before they are split into several sub-samples for analysis with different techniques. A method has been developed whereby dry samples such as sand are homogenized in a shaking machine for one minute before they are split into several sub-samples. Wet solid samples such as soil are homogenized in a mortar for three minutes. Using these methods, satisfactory standard deviations were obtained for the content of DIPMP in the sub-samples.

An evaluation of different solvents for extracting selected chemical warfare agents from the sample materials sand, soil, butyl rubber, and paper showed that a mixture of methanol (50%) and chloroform (50%) gave the highest extraction yields. A number of other solvents also gave high recovery rates.

SAMPLE DOCUMENTATION. SAMPLING.	Page 1 of 3
Items 1-11 refer to the sampling procedure.	
1. Name : Date : Time : Position:	
2. Reason for sampling :	
3. Sample priority : First Second Third	
4. Sample code number :	d
a	e
b	f
c	g
5. Description of sample :	d
a	e
b	f
c	g
6. Method of obtaining the sample(s) :	d
a	e
b	f
c	g
7. Results from field analysis (CAM, TLC) :	
8. Weather conditions :	
9. Description of sample site (topography, vegetation etc) :	
10. Location of sample site (map reference UTM) :	

SAMPLE DOCUMENTATION. SAMPLING. Page 2 of 3

11. Sketch of site , topography, bomb craters etc.
Use cross and sample code number to give sample site.
Please indicate North-South and wind direction.

SAMPLE DOCUMENTATION. ATTACK.	Page 3 of 3
Items 12-17 refer to observations concerning the attack.	
12. Date :	Time :
13. Weather conditions since the attack :	
14. Symptoms of casualties (humans/animals) :	
15. Effect on vegetation :	
16. Description of the attack method of delivery, smell, sound etc)	
17. Description of munitions found at the site :	

18. Remarks :

TRANSPORT LOG.

1. Sampling carried out by : _____ _____

   Confirmed by : _____ _____

   Date : _____ Time : _____ Parcel number : _____

   Total number of parcels in shipment : _____

   Temp should not exceed : _____°C Description of parcel :

2. Transport and storage information.

Date and time received	Place received	Means of transport	Temperature during transport	Name and position

3. Received by _____

   Institution _____ Date : _____ Time : _____

CONFERENCE ON DISARMAMENT

CD/1079
3 June 1991

ENGLISH
Original: FRENCH

LETTER DATED 3 JUNE 1991 FROM THE REPRESENTATIVE OF FRANCE ADDRESSED TO THE PRESIDENT OF THE CONFERENCE ON DISARMAMENT TRANSMITTING THE TEXT OF THE ARMS CONTROL AND DISARMAMENT PLAN SUBMITTED BY FRANCE ON 3 JUNE 1991

I have the honour to send you herewith the text of the arms control and disarmament plan submitted by France on 3 June 1991.

I would be grateful if you would make the necessary arrangements to publish this text as an official document of the Conference on Disarmament and to ensure that it is circulated to all Member States and non-Member States participating in the work of the Conference.

(Signed) Gérard Errera

Ambassador
Representative of France
to the Conference on Disarmament

GE.91-61058/0356H

ARMS CONTROL AND DISARMAMENT PLAN

The spread of increasingly costly and destructive weapons systems is exposing the world to growing dangers. Inequalities, injustices and the imbalance of forces are conducive to threats, the arms race, and conflicts of all kinds. The proliferation of weapons of mass destruction gives a global dimension to these dangers. The whole world, South and North alike, has a vital interest in halting this race and in checking this dangerous and ruinous proliferation. This concerns us all.

Our aims are clear. Chemical weapons must be eliminated. Bacteriological weapons must not be produced. Existing nuclear arsenals must be reduced to the lowest level consistent with the maintenance of deterrence. The non-dissemination of nuclear weapons remains an imperative, whereas the use of nuclear energy for peaceful purposes is justified. The dissemination of ballistic technologies must be strictly controlled in so far as they may be used for aggressive aims. On the other hand, there is no reason to prevent cooperation in space for civilian and scientific purposes.

As regards the so-called conventional weapons, a balance of forces should be maintained, or introduced, everywhere, region by region, while respecting the right of all States to security. Arms exports are consequently to be strictly controlled and so conducted as not to contradict this action. France has already made these goals its own, and is prepared to subscribe to any improvement in existing arrangements, to any new international or multilateral commitment based on them. It has already started thinking about this issue with its partners in the European Community.

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Global arms control and disarmament presupposes action at three levels: the weapons category level, and the regional and worldwide levels.

I. CATEGORY OBJECTIVES

Each weapons category requires specific treatment, proceeding in stages if necessary. In order to achieve clearly-defined goals binding on all, France, which is constantly concerned to adapt its own regulation, proposes that concrete measures, sector by sector, should soon be taken.

1. **Prohibition and elimination of chemical weapons.** The entire international community, meeting in Paris in January 1989, acknowledged the need to complete the convention banning chemical weapons, as soon as possible. France proposed convening the Ad Hoc Committee on Chemical Weapons in Geneva at ministerial level to conclude the negotiations before the end of the year. It calls on all States to become parties to the Convention from the outset.

2. **Prohibition of bacteriological weapons.** At the Review Conference of the 1972 BW Convention in September of this year, France will propose the addition of a protocol on verification.

3. **Nuclear disarmament** remains an essential goal. France supports the efforts to reduce the nuclear arsenals of the two major Powers. It confirms that it will participate in the process as soon as the conditions it specified in 1983 have been fulfilled.

At the same time, it is important to prevent the proliferation of nuclear weapons beyond the present five nuclear Powers.

France, which is already applying all of the terms of the Non-Proliferation Treaty, has in principle taken the decision to sign it and hopes that all States will accede to it.

4. **The Missile Technology Control Régime** (MTCR), which allows those now in possession of missile know-how to limit the risks of ballistic proliferation, should only be a stage towards a more general agreement, one that is geographically more extensive, better controlled and applicable to all. The agreement would lay down rules promoting civilian cooperation in space, while removing the dangers of the diversion of technology for developing a military ballistic capability. Here again, the aim would be to arrive at a situation where all States wishing to gain access to space for development purposes would cooperate in a framework guaranteeing security.

France further recommends the development of outer-space confidence-building measures in the form of a "code of conduct" for civilian and military satellites.

5. **Conventional weapons.** Conventional weapons exports must not counteract the search, region by region, for a balance of forces at the lowest possible level consistent with the right of each State to security.

The five permanent members of the Security Council - which happen to be the principal producers of conventional weapons - **are to start a round of concertation in the coming weeks aimed at determining rules of restraint.** The process could then be broadened on the occasion of the next session of the General Assembly, with a view to establishing an international register of arms sales to be kept by the Secretary-General of the United Nations. Particular attention will be given, in the course of these discussions, to sales of conventional weapons in parallel markets.

The members of the European Community have already begun a process of concertation aimed at making proposals on this subject.

II. OBJECTIVES

The example of Europe is encouraging. The Europeans have moved on from the cold war to peaceful coexistence, and now to cooperation within the framework of the Conference on Security and Cooperation in Europe (CSCE). They have adopted and applied security and confidence-building measures. They have signed the first disarmament agreement concerning conventional weapons. This experience cannot be transposed as such. But it does show that the arms race is not inevitable. This approach could serve as an inspiration for other initiatives in other regions.

We call for **regional security arrangements** based on the following rules:

(a) Only when the solution of conflicts is started, can this process get underway and confidence be created.

(b) Adoption by the parties concerned of **regional confidence-building and security measures.** Information is the prime condition of confidence. Transparency and confidence-building measures, i.e. mutual information on force capabilities and strengths, and on the movements of armed forces, reduce the sense of threat and paves the way for disarmament proper. For that, a framework is needed, which may be a regional organization, and means of inspection in situ and by satellite. France would be willing to disclose information available to it to **regional agencies responsible for transparency.** It would favour the transmission to such regional agencies of the means of observation, in particular those in outer space that as may be available to Europe and the United Nations.

(c) Efforts must be made, through national decisions and through negotiations, to move towards **a balance of regional forces.** Arms export policies must not run counter to this goal. Once this equilibrium has been attained, its level should progressively be lowered through negotiations providing for means of control.

(d) **Strict regional application of the category régimes laid down for nuclear, bacteriological and chemical (NBC) weapons and ballistic missiles.** That is why, at the initiative of France, United Nations Security Council resolution 687 on the cease-fire in the Gulf conflict noted that the specific constraints imposed on Iraq as regards nuclear, chemical and biological weapons, as well as ballistic missiles, represented steps towards the goal of establishing in the Middle East a zone free from weapons of mass destruction and all missiles, along with a global ban on chemical weapons. That goal should be pursued in other regions of the world.

III. The United Nations is now in a position to fully play the role it was assigned by its Charter. France considers that it is therefore for the **Security Council** to guarantee and, if necessary, to harmonize these disarmament and non-proliferation policies. The Council must encourage the signature of agreements on particular arms categories, as well as regional and multilateral disarmament and non-proliferation agreements. From the agreements reached, it would derive general rules and thus exercise a function of worldwide vigilance.

This presupposes that the members of the Security Council and in the first place its permanent members will set the example.

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CONFERENCE ON DISARMAMENT

CD/1079/Corr.1
11 November 1991

ENGLISH only

LETTER DATED 3 JUNE 1991 FROM THE REPRESENTATIVE OF FRANCE ADDRESSED TO THE PRESIDENT OF THE CONFERENCE ON DISARMAMENT TRANSMITTING THE TEXT OF THE ARMS CONTROL AND DISARMAMENT PLAN SUBMITTED BY FRANCE ON 3 JUNE 1991

Corrigendum

On page 3, the subtitle should read:

"II. REGIONAL OBJECTIVES"

GE.91-62735

CONFERENCE ON DISARMAMENT

CD/1080
CD/CW/WP.341
5 June 1991

Original: ENGLISH

UNITED KINGDOM OF GREAT BRITAIN AND NORTHERN IRELAND

VERIFICATION OF THE CHEMICAL WEAPONS CONVENTION: PRACTICE CHALLENGE INSPECTIONS AT CIVIL CHEMICAL PLANTS

Introduction

1. In CD921 and CD1012 the United Kingdom presented the results of its practice challenge inspection programme at government facilities. Our key conclusion was that there was no site so sensitive that we could not allow some form of access within the site, albeit rigorously managed. We subsequently decided to examine whether the lessons learned during the government programme were also relevant to inspections at civil chemical plants. One of the key issues we wished to examine was how to protect commercial confidentiality whilst still providing sufficient access for the inspectors. Accordingly one full practice challenge inspection at a large multi-purpose batch plant and a follow-up visit to a separate smaller dedicated plant were held in January and April this year. The follow-up visit was used to explore in detail some of the key lessons identified during the practice inspection. This visit reinforced many of the principal conclusions drawn in the full inspection.

Aims and Objectives

2. The inspection and follow-up visit had two major objectives:

(i) to test the effectiveness of challenge inspection at chemical plants, especially at large multi-purpose facilities.

(ii) to see whether sufficient access within the plant and to documentation could be given to demonstrate compliance without seriously compromising commercial confidentiality.

Conduct of the Exercise and Major Lessons

3. The first inspection involved an inspection team of 5 inspectors; a sampling team of 3; a receiving team of 9, including both government and company personnel; 2 control staff and one observer. For the purposes of the trial, it was agreed in advance to limit the inspection to one production area only within the large multi-purpose facility. This still provided a representative indication of the problems likely to be encountered in a real inspection, and contained sufficient interest to sustain two days of inspection. The follow-up visit focussed in greater depth on the scope for using a dedicated plant to make alternative products, the scope for diverting production, and examination of production, health, and safety records.

4. The civil inspection exercise reinforced many of the organisational and practical lessons learned during the government practice challenge inspection programme: for example, the importance of relevant expertise in both inspection and receiving teams; the requirement for well worked out sampling procedures and the need for clear rules for the challenging state observer (CSO). In the course of the inspection several valuable lessons were learned about the handling of challenge inspections at civil chemical plants. These can be summarised under the following headings: compliance assessment; records; commercial confidentiality; sampling practicalities; government expertise; and financial aspects. These issues are discussed in fuller detail below.

Compliance Assessment

5. From the inspectors' point of view, a challenge inspection at a chemical plant can present a more difficult task than at a military facility for the simple reason that the inspectors are "surrounded by chemistry". Inspectors therefore need to ensure particular precision in gathering information on chemicals present at the site, and this may require a more detailed analysis of the plant's activities to provide sufficient confidence in compliance.

6. If the request for an inspection does not give precise details of the compliance concern, the inspection team would have to rely on their own judgement in testing for particular Schedule 1 chemicals or other potential or known CW agents. In general an inspector's tasks might include the following:

- check for evidence of the presence of Schedule 1 chemicals on site shortly before the inspection;

- check that there are no chemicals on the site at the time of the inspection which are in contravention of the CWC, or inconsistent with declarations;

- check that any chemicals with CW potential are in fact being used for legitimate purposes and are not being diverted;

- look for evidence of possible violations in the recent past.

7. If a plant was designed with nerve agent production in mind, and was readily adaptable, this would probably be picked up quite easily by the inspectors, particularly if key precursors were present on site. Other CW agent production might be harder to detect. The inspection team would need to include at least one chemical process engineer familiar with requirements for classical CW agent production. Inspectors would also need to be aware of possible requirements for production of "novel agents". The inspectors could then target their efforts accordingly. A precise specification of the compliance concern in the inspection request eg "believe production of GB or GF occurring in..." would considerably facilitate inspection and analytical tasks, particularly at large complex chemical plants. It would also reduce the risks to commercial confidentiality since a less intrusive inspection may be required to satisfy the particular compliance concern in such circumstances.

8. The inspectors would have required a full 7 days to undertake an effective inspection of the whole of the large and complex chemical site which was the subject of the first UK trial. Despite potential practical and evidential difficulties we concluded that an inspection would have an important deterrent value. The inspected State would not know exactly which plant, and in what sort of detail, the inspectors would chose to examine over the 7 day period. Nor, moreover, could the challenged State be confident that all traces of potentially incriminating evidence could be completely removed.

Records

9. At the UK exercises, the inspectors did not, overall, find commercial confidentiality requirements a major obstacle to their assessment of the site's activities. The only problem of note they encountered concerned the analysis of records for a particular Schedule 3 chemical where the receiving team were initially prepared to offer only documentation from computer records to show inspectors that all materials were used as stated. This was regarded by the inspectors as of limited value in building confidence. Computer printouts, especially when produced in a location physically separate from the actual production line, do not have sufficient verisimilitude to provide convincing evidence. Sight of original hand-written records and invoices for the delivery of relevant chemicals would add considerably to the credibility of computer records. In the case mentioned above it proved possible to demonstrate a plausible internal consistency for randomly selected month, week and batch numbers, having been given some information on the raw materials for the product in question.

10. The effort involved in maintaining a second, false, set of records consistent with the scale of operations on site and internally consistent throughout would be considerable. Diversion of small percentages of chemicals could, however, be hidden within apparently legitimate mass balances. This suggests that using record analysis to verify such consistency, rather than for determining mass balances, may be a more effective use of inspection time and play a valuable role in verifying compliance at a chemical plant. To this end, the inspectors should consider examining laboratory quality control, maintenance, health and safety records, as well as production records. Some familiarity with industrial record keeping systems among the inspection team would obviously be helpful here. Access to sections of the overall records for periods in time selected by the inspection team would limit the amount of commercially sensitive information that needed to be divulged. But the ability to authenticate particular records by cross-checking throughout can provide greater confidence to the inspection team. The more transparent individual companies can be, the easier it is to demonstrate that all activities are indeed legitimate.

Commercial Confidentiality

11. Some attention has already been given in the negotiations to the question of protecting legitimate commercial secrets in the development of the Annex on the Protection of Confidential Information. In practice, as was evident in the UK exercises, private companies will naturally remain concerned not to reveal, even to the inspection team, any more confidential information than is necessary for the purposes of the inspection. They may be particularly concerned at the steady accumulation of information during the course of an inspection. Uncertainty on

the part of individuals at a plant as to how much information to reveal could lead to over-caution in responding to inspectors' questions. One way to address this problem might be for one or possibly a small core team of senior plant representatives to accompany the inspection team so that all questions and answers can be channelled through them. Individual companies might also consider preparing a general guidance note for all their manufacturing sites so that they can prepare, if necessary, relevant outline plans for receiving an inspection. Where there are particular concerns, companies could identify beforehand, as far as practicable, the most sensitive information and processes which they would prefer to protect, and consider how they would provide reassurance to inspectors if pressed on these points. Such plans might need to be updated from time to time to take account of any changes that may occur at the site. In practice general guidance may be all that is feasible or indeed necessary.

Sampling

12. Not surprisingly, sampling at the chemical plants proved to be a more complex activity than at our previous inspections at non-chemical sites. Industrial safety procedures for sampling hazardous materials were extremely stringent and required proper certification and the use of trained site personnel. In addition modern chemical plants may have long complicated pipe runs and not be readily susceptible to sampling at multiple points. In the first UK exercise the inspection team used process flow charts to follow through the production line of one product in order to identify the optimum sampling locations to verify that production was as claimed and that no controlled

chemicals were present. The company was particularly concerned about two of the chosen locations, fearing that detailed analysis would reveal sensitive data eg knowledge of reaction components which, if leaked, could provide competitors with details of reaction mechanisms. The inspection team overcame this problem by allowing the company to perform analysis in its own laboratories under their close supervision.

13. Since many classical CW agents contain phosphorus and sulphur, gas cromatography with flame photometric detection could be used. Alternatively, the use of Neutron Activation Analysis could be employed by the inspectors themselves. Both techniques could probably provide sufficient information to satisfy the inspectors without having to divulge commercial secrets. The method of element specific analysis, although widely applicable, may not work in every case, but the principle of negotiating restricted analytical methods can provide a valid approach to dealing with confidentiality concerns and might in some cases avoid the need for off-site analysis. In the UK exercise the company was able to find a way of satisfying the inspectors while keeping commercially sensitive information secret.

14. Six main conclusions were drawn on the practical aspects of sampling at chemical plants. These were:

- internationally acceptable safety certificates determining the intrinsic safety of electrical equipment would simplify and expedite home team clearance of approved items;

- inclusion in inspection teams and sampling teams of personnel fully conversant with the safety equipment (eg CCBA suits) necessary to enter particular plants would reduce or even preclude the need for in situ training and avoid opportunities for obstruction and delay on the part of the home team. This might require international agreement on universal standards;

- availability of process flow diagrams would allow sampling to be more specific and so reduce the numbers of samples that need to be taken;

- instruments, such as gravity meters, to assist in detecting underground systems would be useful;

- additional aids to sampling such as long handled dippers, extending vapour samplers etc (for vent sampling) would make the process easier at many locations within a chemical plant;

- sampling can be time-consuming and requires a team of at least 3 members for the task to be performed effectively and efficiently.

Financial Aspects

15. The direct costs of the inspection in terms of the provision of facilities were relatively minor, although there were additional resource implications in the involvement of company personnel. The key financial concern from the company's point of view is to ensure that their production and delivery schedule is not adversely affected since this can have potentially damaging economic consequences. As in the case of

IAEA safeguards, it needs to be stressed that inspection activities should not unduly interfere with legitimate production at civil chemical plants. At the same time it will be in the company's interests to be ready to propose ways of addressing the inspectors' concerns which minimise the impact on their legitimate activities eg sampling from adjacent areas, process flow diagrams, access to plant control instrumentation, waste outflows or treatment plants, feed stocks and filling points. Similarly, to enable the challenged site to be secured during the inspection whilst allowing legitimate activities to continue, the inspection team could allow traffic to move in and out of the site but subject it to random checking. This would give confidence that no CW-related material is being removed whilst not disrupting manufacturing and delivery schedules.

Government Expertise

16. Our practice challenge inspection programme at government facilities demonstrated the importance of government providing technical and policy advice and guidance for each challenged facility. Our recent trials showed this to be true also for civil chemical plants. We would envisage having a senior policy or military official to lead the home team and be the main point of contact with the inspectors rather than placing this burden on a site superintendant or plant manager. A national implementation committee might also be necessary to provide extra personnel and administrative support for escort duties and transport to reduce the burden placed on individual companies. Government expertise and advice on handling challenge inspections would need to be made generally available to the civil chemical industry. Chemical firms might wish to consider training designated personnel in the management and conduct of inspections. Practical experience gained from trial inspections should prove very valuable in preparing implementation procedures, particularly at sensitive sites.

Conclusions

17. Although the civil inspection programme was not as extensive as the trials at government facilities, the combination of a full practice challenge inspection at a large complex chemical plant and a follow-up visit to a smaller dedicated facility provided a useful insight into some specific aspects of handling challenge inspections at civil chemical sites and enabled some valuable lessons to be drawn. Our main conclusions were as follows:

(i) Concerns Over Commercial Confidentiality

The need to protect legitimate commercial confidentiality can be reconciled with an inspection team's requirements for extensive access. The duty of an inspection team not to interfere unreasonably with the legitimate work of a company should be spelt out in greater detail in the Inspection Protocol. If a stoppage of production is requested, companies should be ready to explain their difficulties and suggest how these might be overcome.

The chemical complexity of some sites and the size of some companies may make it difficult for companies to prepare thoroughly in advance for a possible inspection, but companies may find it useful to provide general guidance to their staff and to select local staff to receive and accompany any inspection team. Help in preparing for and receiving an inspection should be available from a government team of experts.

(ii) Sampling

The inspectors will need to be accompanied by a full team of properly trained and well equipped experts to carry out sampling. A system of international safety clearances, which the samplers would need to satisfy, would avoid repetition of safety training at each site. Element specific analysis and giving companies the opportunity to carry out analysis of samples themselves where possible, under the guidance of the inspectors, would help to protect confidentiality.

(iii) Detection of Non-Compliance

Although breach of the Convention might be more difficult to detect at a chemical site than at a military site, a wide ranging inspection based on examination of plant equipment and checking of records would present a significant risk of discovery to an evader.

CONFERENCE ON DISARMAMENT

CD/1081
CD/NTB/WP.13
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Original: ENGLISH

AUSTRALIA AND NEW ZEALAND

VERIFICATION OF A COMPREHENSIVE TEST BAN

Summary

New Zealand and Australia share a common commitment to the achievement of a comprehensive ban on nuclear testing. The resolution submitted jointly by Australia and New Zealand to last year's General Assembly emphasises that:

> "An end to nuclear testing by all states in all environments for all time is an essential step in order to prevent the qualitative improvement and development of nuclear weapons and their further proliferation, and to contribute, along with other concurrent efforts to reduce nuclear arms, to the eventual elimination of nuclear weapons".

2 Despite overwhelming endorsement in the United Nations of the call for the early achievement of a CTB, there has been little substantive progress towards this goal. New Zealand and Australia share the sense of frustration felt by many countries that the major achievements of recent years in disarmament and arms control have not been matched by comparable progress on a nuclear test ban. The two countries saw the Partial Test Ban Treaty Amendment Conference as a good opportunity to discuss in depth ways of furthering the objective of a CTBT, particularly on the important issue of verification. This paper was originally submitted jointly to the Conference as a positive and constructive contribution to its discussions.

3 The achievement of an effective CTBT is of course a global concern, and the need for verification clearly is not confined to the existing nuclear weapons states. New Zealand and Australia firmly believe that a CTBT has an essential role in preventing the horizontal and vertical proliferation of nuclear weapons. The achievement of a verifiable nuclear test ban would considerably strengthen the non-proliferation regime based on the NPT, IAEA safeguards and nuclear free zones.

Verification Requirements

4 One fundamental argument that has been put forward against a CTBT is that a nuclear test ban would not be verifiable. The Governments of Australia and New Zealand do not accept this view. Substantial scientific evidence is now available establishing the feasibility of verifying a nuclear test ban, particularly through seismic monitoring. It is technically possible to detect underground nuclear tests with a high degree of confidence down to very low levels.

5 In the view of New Zealand and Australia the development of an effective verification mechanism is an important part of the process of creating an effective, widely supported, and verifiable CTBT. Effective verification is however only part of the whole process which will need to involve a political will on the part of a number of key players before it can be achieved. In the meantime we see great value in taking as far as we can the development of an effective verification mechanism.

Seismic Verification Techniques

6 The two countries consider that the work carried out within the Conference on Disarmament by the ad hoc Group of Scientific Experts (GSE) is of fundamental importance in demonstrating that technical means exist for the verification of a CTBT.

7 The first report of the ad hoc group in 1978 (CCD/558) presented the results of a study of the detection ability of a hypothetical global network (Network III (SRO)) of digital broad-band seismographs located at then existing seismograph stations. It was estimated that this network (of at least two sections) would have a 90 per cent chance of detecting an event of magnitude 3.8 to 4.2 (or greater) in the northern hemisphere and magnitude 4.0 to 4.6 (or greater) in the southern hemisphere. Magnitude 4.0 corresponds to an explosion of less than ten kilotons in most geological environments and much less than this level in some environments.

8 The third report of the group (CD/448) in 1984 pointed to significant developments in seismic instrumentation, including the installation of more seismic arrays. Since that date, there have been still greater improvements, particularly in response to the initiatives of the ad hoc group. While further study is needed to evaluate accurately the capability of the present global network, it is clearly very close to that projected in 1978, and may well be significantly better.

GSETT-2

9 New Zealand and Australia both strongly support, and are participating in, the second technical test on the global exchange and analysis of seismic data being conducted by the group (GSETT-2). Phase 3 of this test involving world wide data exchanges for a period of 42 consecutive days has just concluded. The two countries have contributed data from monitoring stations in Australia and New Zealand, and also from stations as far apart as the Cook Islands and Antarctica. All together, this seismic monitoring system covers a major part of the southern hemisphere, and represents an essential component in the global scope of the experiment.

10 Detailed evaluation of the test, which will take place in Phase 4 and be presented in the group's report, will be needed to assess the extent to which global seismicity has been monitored. However, it is already clear that from an operational point of view the test has been very successful. Modern communications systems have provided efficient means of data transmission from stations to international centres. At these centres, large computing facilities accomplished the huge task of assembling the mass of data pouring in from the network, and daily bulletins incorporating such data have generally been available within seven days. Requests from individual states for data to assist their national verification studies were responded to promptly by the centres. It is clear that the global system established for GSETT could well provide the basis for an effective system under a comprehensive test ban treaty.

11 Both New Zealand and Australia have recently upgraded their seismic monitoring and data transmission equipment as a further part of their contribution to GSETT-2. Australia acted as one of the four international data centres for this experiment. The seismic arrays operated by Australia have the facility for enhancing weak signals by data processing techniques, which makes a major contribution to the global network's detection ability, particularly in the southern hemisphere.

12 The two countries were well placed to contribute to the experiment because of their extensive experience in using seismic detection to monitor nuclear tests. Both New Zealand and Australia have for many years been closely monitoring France's nuclear testing programme at Mururoa Atoll in the South Pacific. Scientists in New Zealand and Australia have accordingly developed considerable practical expertise in the seismic monitoring of underground nuclear tests of varying magnitude.

13 New Zealand's monitoring of explosions in French Polynesia, using a seismograph station in the Cook Islands, has demonstrated the particular sensitivity of such stations to tests in oceanic areas. This would be an important factor in a global monitoring network.

14 Based on this collective experience, it is the considered view of the Australian and New Zealand Governments that seismic monitoring offers a reliable means of verifying a CTBT. The successful conclusion of GSETT-2 will further strengthen confidence in the effectiveness of a global seismic monitoring and data transmission system down to a very low level.

Evasion

15 The possibility of evasion has been put forward as a reason to doubt that a CTBT could be adequately monitored. Various technical scenarios have been put forward for ways in which countries might evade detection, primarily through 'decoupled' explosions in underground cavities. However, the technology to attempt such evasion is unlikely to be available outside the existing nuclear weapons states, and the risk of detection would be extremely high and would increase with each test.

16 With in-country networks of seismographs to enhance the detection capability of the global network, it is likely that even the most elaborate precautions would not prevent the detection of explosions at levels as low as 1 kiloton. Attempts to evade a nuclear test ban would therefore be confined to a level at which the military advantages to be gained from clandestine explosions would be minimal and the chances of escaping detection would be extremely low.

Other Verification Techniques

17 While seismic detection would be a vital factor in the monitoring of a CTBT, verification need not depend on this alone. A network for monitoring airborne radiation, as proposed by Sweden, would provide valuable collateral support to a global seismic monitoring network. Satellite surveillance data would also help confirm international adherence to a CTBT.

18 A recent development has been the increased acceptance of on-site verification in other areas of arms control, following the precedent first established by the

INF Treaty. This could also be extended to verification of a nuclear test ban. The ratification of the protocols to the Threshold Test Ban Treaty and the Peaceful Nuclear Explosions Treaty has now set a further example for cooperation on verification procedures.

19 On-site verification, which is already widely accepted as an important component of arms control agreements, might also provide further assurance of compliance with a CTBT. The international disarmament process has advanced dramatically since the seismic monitoring network was first proposed, when national technical means were considered the only effective means of verifying compliance. Cooperation on measures for on-site verification, such as the placement of seismometers at possible test sites, would strongly reinforce a teleseismic verification system.

Wider Participation in Seismic Cooperation

20 In order to develop a global seismic monitoring capability down to a very low level it will be necessary to improve the coverage of seismic stations in Africa, South America and Antarctica. The involvement is therefore necessary of a wider group than is currently involved in the group of scientific experts (GSE) in the Conference on Disarmament.

Conclusions

21 In summary, New Zealand and Australia consider that there already exists compelling scientific evidence that a nuclear test ban is verifiable using technology currently available. The ongoing work of the Group of Scientific Experts is providing substantial technical corroboration for that view. The argument advanced by some nuclear testing states, that a nuclear test ban is not possible because it could not be verified, has consequently become increasingly difficult to sustain.

22 New Zealand and Australia consider that considerable progress has already been made towards the development of an adequate mechanism to verify a nuclear test ban. The ongoing work of the Group of Scientific Experts is important in this respect and it needs to continue its work with the benefit of a wider membership.

23 New Zealand and Australia welcomed the constructive discussion of verification issues at the Partial Test Ban Treaty Amendment Conference. The two countries had suggested that a significant achievement for the conference would be to:

- recognise the progress made towards a fully effective international verification system;

- endorse the efforts of the Group of Scientific Experts to demonstrate the technical feasibility of such a regime;

- call on more states to participate in the GSE to improve in particular coverage of seismic stations in Africa, South America, and Antarctica.

The widespread support for these general principles was encouraging. New Zealand and Australia believe that as a result of the discussions in New York there is greater international commitment to the important work on verification being carried out by the Group of Scientific Experts under the auspices of the Conference on Disarmament.

CONFERENCE ON DISARMAMENT

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SPAIN

REPORT ON A NATIONAL TRIAL INSPECTION IN THE CIVIL CHEMICAL INDUSTRY

INTRODUCTION

The Spanish Government has repeatedly emphasized its resolve to give firm support to a convention banning chemical weapons which will be effective and consequently verifiable.

In that context, and with the aim of bringing its own experience to bear on the negotiations and the objectives of the convention, Spain carried out an inspection of a plant producing a range of products, in which a schedule 3 chemical is produced by industrial chemical processes.

The inspection procedures applied were those stipulated in the text of the draft convention for schedule 2 chemicals.

The data obtained from the national trial inspection are presented in accordance with the now standard model set out in document CD/CW/WP.213.

A. GENERAL APPROACH

1. Objectives of the national trial inspection

The national trial inspection had the following objectives:

To assess the difficulties arising in practical application of the procedures laid down for verification of chemical industry producing schedule 2 chemicals;

To assess the relations between inspection of the chemical and industrial aspects of the plant and inspection of the accounts (auditing), using documents supplied by the management.

The trial inspection also had the following indirect objectives:

To provide direct information and experience to one of the seven major chemical plants in Spain, which, like all the others, will be affected by the provisions of the chemical weapons convention;

To provide direct information and experience to agencies in the Spanish administration which in the future will also be involved in tasks related to the application of the chemical weapons convention, both from the viewpoint of internal policy (new laws and regulations) and from the viewpoint of relations with the bodies to be set up under the convention (national authority, experts working for the organization, and so on).

The experience acquired proved particularly useful for a country like Spain, which has a large chemical industry with a medium-to-high level of technological development and a substantial export capacity.

2. Provisions in the draft convention under which the trial inspection took place

The inspection was conducted in accordance with the rules laid down for verification and monitoring of schedule 2 chemicals appearing in annex 2 of article VI. Despite the fact that the inspected plant does not produce schedule 2 chemicals, it was selected because its major features are very similar to those which would have to be declared under annex 2 of article VI.

3. Type of on-site inspection

The national trial inspection involved an initial, general inspection of the plant for familiarization purposes, followed by a routine on-site inspection.

4. Advance information

(a) Declarations

The plant management provided the inspectors with a detailed declaration in accordance with the format set out in annex 2 of article VI.

(b) Agreement on inspection procedures

The inspection programme was drawn up at a meeting held by the inspectors with representatives of the company one week before the inspection took place.

5. Type of facility inspected

The inspected plant corresponded to a declared multipurpose facility whose principal product is basic monomers for plastics production.

6. Type of declared activity at the facility

The plant produces no chemicals listed in schedule 2 of the draft convention text. However, it does produce a chemical on schedule 3, which, for the purposes of the inspection, was considered to be one of those included in schedule 2.

The plant consists of three clearly separate parts:

In the first, two conventional feedstocks A and B react over a catalyst in a fluidized bed to produce chemical C, the principal product of the reaction, and as a by-product chemical D, which was the principal subject of the inspection.

As is normal with organic chemistry reactions, a number of waste products are also produced which are not commercially usable and which are subsequently sent with others for destruction by incineration in a purpose-built unit within the plant itself.

The reaction liquid is subject to a process of separation by distillation as a result of which chemical C, following purification, is ready for sale.

Chemical D is used primarily to produce a further chemical E, an intermediate product normally traded in the chemical industry.

Another portion of chemical D, which was the subject of the inspection, is made into an aqueous solution which is transported by rail to an organic chemical plant elsewhere in Spain.

This is the real crux of the inspection, since at this point chemical D has not been processed by the producer. However, the inspection team noted that for security and environmental reasons, the competent authorities at the time had not authorized the installation of intermediate tanks, so that there is no more chemical in the plant than that found in the container in which the solution is prepared, and that, once it is full, this container is pumped out into a tank wagon for transport to its destination.

Subsequent visual inspection confirmed that the facility contains no equipment for the compression, liquefaction, packaging or transport of this chemical in another form.

7. Actual activity at the facility

B. DETAILED APPROACH

1. The inspection agreement

The management of the company and the inspection team negotiated a very broad inspection agreement under which there were no restrictions on access to any part of the plant. The agreement was drawn up on the basis of the document set out in CD/1033.

2. The inspection team was made up of the following:

Team leader: A chemical industry manager with extensive experience in industrial processes;

A chemical plant manager who is an expert in processes, with special reference to security and environmental factors;

A weapons specialist representing the Ministry of Defence;

A chemical warfare specialist representing the Ministry of Defence;

An expert in commercial and balance-sheet auditing;

Two representatives of the Ministry of Foreign Affairs, performing functions which will in future be allocated to the national authority.

The inspectors were accompanied by a representative of the company that owns the facility visited.

3. Inspection equipment

During the inspection, in compliance with applicable regulations, use was made of protective security equipment supplied by the company itself. Equipment for sample-taking and analysis was also supplied by the company and used on the spot.

4. Activities prior to the arrival of the inspection team

The plant management was given one week's notice of the exact date of the inspection.

5. Advance preparation on-site

The only preparations carried out on-site were those immediately connected with the work of the inspection team. Consequently, the plant made no changes in its normal procedures in advance of the inspectors' visit.

6. Preparations relating to escort and points of contact

The inspectors were accompanied by staff of the company which owned the facility, starting from a point of contact decided upon by the company itself.

7. Other participants

There were no other participants.

8. Duration of inspection

The inspection took three days in all. On the first day the first contact was made with plant representatives and the confidentiality agreement was signed. On the second day the inspection team split into two groups; the scientific/chemical group began the detailed inspection of the plant after receiving precise information from the technical department.

The accounting expert visited the accounting department and began his study of the documentation supplied by the company.

The inspection team devoted the third day to drafting its report on the basis of the information obtained by the two groups of inspectors in their fields of competence.

9. Measures to protect confidential information

Prior to any activities related to the inspection, the plant management required all the members of the team to sign a confidentiality agreement in which they undertook not to use the information obtained as a result of the exercise for purposes unrelated to the inspection.

This undertaking is binding unless waived by the company itself.

10. Opening conference

During the opening conference, company representatives elaborated in detail on the preliminary information provided one week before and explained the safety regulations that would have to be complied with in the course of the inspection.

The company's Processes Department presented production data for the previous two years, the flow chart and the manufacturing process for the product under investigation.

11. Types of records needed and/or audited

One member of the inspection team devoted himself to the authoritative verification, on the basis of the commercial documentation on inputs and outputs, of the data and flow charts supplied. It should be noted that this company's operations are audited by firms of recognized international competence.

The firm's balance sheets at the beginning and end of the past few years were examined with respect to stocks of those products used in the production processes of the units examined.

By means of the relevant company documents, production for these years was studied and checked, together with other inputs from units other than those involved in the process in question.

The records of intermediate products dispatched to other units, internal consumption and commercial sales were then examined.

On completion, these balances for the whole production process were compared with the figures obtained by applying efficiency factors to each unit in the light of the relevant levels of use; it was found that the recorded and calculated product balances matched perfectly.

After these products had been matched, the team used random sampling methods to analyse invoices and records of purchases of raw materials, transfers of intermediate products and sales of commercial products.

Lastly, the team checked all the supporting documents relating to sales of the product with which the inspection was directly and specifically concerned, and found that they were in complete conformity with the sales data provided by the company in its product balance.

Full monitoring of product D could, if necessary, be achieved by comparing shipping documentation with the acceptance documentation of the receiving company, and comparing both sets of documentation with that of the Spanish National Railways, since products of this type are carried exclusively by rail.

12. Plant orientation tour

After the analysis of the plant's characteristics and flows, and of all the information made available by the management after the opening conference, the team made a visual inspection of the plant which from the outset confirmed the data it had already received.

13. Inspection of areas and facility equipment

The following areas and facility equipment were inspected in detail:

Reaction area;

Repairs area;

Purification area;

Distillation area;

Storage area;

Laboratories, sample-taking processes and analytical equipment.

14. Inspection of operation procedures

In the course of the inspection, special attention was paid to those procedures used in the production of the chemical under inspection in which diversion to the manufacture of schedule 1 substances might occur.

15. Sampling and sample-taking procedures

Sampling at the various stages of the production process and sampling from waste were undertaken by company personnel using company equipment whenever and wherever the inspectors so requested.

16. Handling of samples

The samples were always taken under the direct supervision of the inspectors, and the sample containers were identified by the inspectors, in order to safeguard subsequent handling.

17. Analysis of samples

The samples obtained throughout the production process under the supervision of the inspectors were immediately analysed in the plant laboratory, again under direct supervision by the inspectors. The laboratory log-books recording the systematic analyses of the aqueous solution of substance D were also examined and were found to conform to the analyses made by the company's clients who had received the substance.

18. Types of analyses

The analyses were carried out in conformity with ASTM standards.

19. Documentation of the inspection

The inspectors kept records of the samples taken throughout the production process and of the results of the analyses carried out immediately afterwards in the plant's own laboratory.

It was not considered necessary to remove any documentation from the plant.

20. Evaluation by inspectors

The evaluation undertaken by the inspectors took into account:

The complete documentation provided by the company as regards technical accounting data;

The possibilities of detecting anomalies in the course of the production process;

Consistency between inspection of the production plant and the accounting data supplied;

The firm's willingness to cooperate with the inspectors.

21. Closing conference

No conference was necessary on completion of the inspection team's activities.

22. Anomalies, disputes and complications

None.

23. Report of the inspection team

The report on the inspection was prepared in the course of a meeting held immediately after the inspection and was drafted by one of the team members during the days following that meeting. It comprises the following sections:

Preparations for the inspection: prior contacts and conversations;

Analysis of the confidentiality agreement;

List of inspection activities undertaken at the plant;

Analysis of accounting and commercial documents provided by the company;

Results and verification of analyses and analytical systems;

General evaluation and conclusions by the inspectors.

24. Impact of the inspection on facility operations

The chemical industrial and accounting aspects of the trial inspection caused no disruption of the plant's production process.

Nevertheless, the inspection did directly entail expenditure for the company in the form of loss of working hours by staff and travel by some of its executive staff both during the preparatory period and during the inspection itself.

C. CONCLUSIONS

1. The inspection mandate

The inspection mandate was sufficiently broad to enable the inspection team to do its work effectively. The work was effective in that it enabled reasonable certainty to be achieved concerning the plant's activities. The company denied the team access only to the computerized data bank in which the plant's production processes are stored. Apart from this type of information, which lies at the heart of the company's legitimate right to confidentiality concerning its technological and commercial research, the inspectors had free access to all departments.

2. Composition of the inspection team

Inspection of a multi-purpose plant of the kind with which we are concerned requires a large inspection team, as experience has shown. The inspection team must comprise experts in chemical engineering, chemical analysis and commercial and balance-sheet auditing. This obviously entails costs which will probably in some way devolve upon the chemical industry

inspection systems that are eventually adopted under the chemical weapons convention. Moreover, and in contrast to the experience of other countries, after trial inspections such as this it does not seem that a smaller plant can be inspected by a smaller team of inspectors. Because of the complexity of current industrial processes, for the purposes of in-depth analysis a high and specialized degree of technical expertise is required, whether the plant be large or small.

It was also found that the work of the accountants as team members was fundamental. As in the case of other countries, the Spanish experience has shown that the modern industrial chemical process generates a large quantity of records, the analysis of which is of great benefit to the inspectors in their work. Moreover, in the majority of cases, this analysis does not involve serious interference in the company's privacy (raw material inputs, movements of substances within the plant, etc.).

3. Analysis and taking of samples

The systems used for the taking of samples and immediate analysis of the samples obtained whenever and wherever the inspectors so requested enabled them to ascertain to their full satisfaction the legitimacy of the plant's activities. It is, however, more than likely that on many occasions, for purely technical reasons, it will be necessary to take more samples and to subject them to comparative analysis within and outside the plant, using procedures that are much more stringent and are clearly separate from the channels, procedures, facilities and even personnel of the plant.

4. Duration of the inspection

In the opinion of the team of inspectors, the time taken up by the inspection (three days on-site, plus a preliminary meeting one week before) represents a realistic estimate of the possible duration of an actual inspection in the future.

5. Measures to protect confidential information

From the outset of its contacts with the inspection team, the company expressed its concern about confidentiality. Consequently, all members of the team were required to sign a document in which they undertook not to divulge any of the data obtained in connection with their activities. It was not considered feasible to draw a distinction between confidential and non-confidential data.

6. General assessment

The inspectors realized the importance of good cooperation on the part of the company, both in the preparation and in the conduct of the inspection. They also concluded that no firm whose activities are legitimate would be likely to have a serious objection to supplying the inspectors with exhaustive documentation on technical data relating to the facility, production charts, efficiency levels and analytical methods.

The inspection team also realized the enormous importance of visual inspection - both prior overall inspection and subsequent detailed inspection. Through careful visual inspection unrestricted by the company, an expert can already obtain important information.

Lastly, it was found that commercial auditing, especially in those companies in which the documentation used conforms to examinations by internationally recognized external auditing firms, is vital and ensures reliable checking of figures and activities. The use of this instrument for this type of verification is accordingly considered highly advisable.